

(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau(43) International Publication Date
18 December 2003 (18.12.2003)

PCT

(10) International Publication Number
WO 03/104255 A2

- (51) International Patent Classification⁷: C07K (74) Agents: RICHARDS, John et al.; Ladas & Parry, 26 West 61st Street, New York, NY 10023 (US).
- (21) International Application Number: PCT/US02/33603 (81) Designated States (*national*): AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, OM, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW.
- (22) International Filing Date: 21 October 2002 (21.10.2002)
- (25) Filing Language: English
- (26) Publication Language: English
- (30) Priority Data: 60/348,974 23 October 2001 (23.10.2001) US (84) Designated States (*regional*): ARIPO patent (GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, SK, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).
- (71) Applicant (*for all designated States except US*): NEURO-GEN CORPORATION [US/US]; 35 Northeast Industrial Road, Branford, CT 06405 (US).
- (72) Inventors; and
- (75) Inventors/Applicants (*for US only*): BLUM, Charles, A. [US/US]; 785 W. Pond Meadow Road, Westbrook, CT 06498 (US). BRIELMANN, Harry, L. [US/US]; 14 Elm Street, Guilford, CT 06437 (US). DE LOMBAERT, Stephane [BE/US]; 37 Concord Drive, Madison, CT 06443 (US). ZHENG, Xiaozhang [CN/US]; 10 Roby Court, Branford, CT 06405 (US).
- Published:
— without international search report and to be republished upon receipt of that report
- For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.*

WO 03/104255 A2

(54) Title: SUBSTITUTED 2-CYCLOHEXYL-4-PHENYL-1H-IMIDAZOLE DERIVATIVES

(57) Abstract: Substituted 2-cyclohexyl-4-phenyl-1H-imidazole derivatives capable of modulating NPY5 receptor activity, are provided. Such compounds may be used to modulate NPY binding to NPY5 receptors *in vivo* or *in vitro*, and are particularly useful in the treatment of a variety of disorders (e.g., eating disorders such as obesity or bulimia, psychiatric disorders, diabetes and cardiovascular disorders such as hypertension) in humans, domesticated companion animals and livestock animals. Pharmaceutical compositions and methods for treating such disorders are provided, as are methods for using such compounds for detecting NPY5 receptors.

SUBSTITUTED 2-CYCLOHEXYL-4-PHENYL-1H-IMIDAZOLE DERIVATIVESField of the invention

This invention relates generally to substituted 2-cyclohexyl-4-phenyl-1H-imidazole derivatives that are modulators of mammalian neuropeptide Y₅ (NPY₅) receptors, and to the use of such compounds for treating a variety of physiological disorders associated with NPY₅ receptor activation, such as feeding disorders, psychiatric disorders and cardiovascular diseases. The invention further relates to the use such compounds as probes for the detection and localization of NPY₅ receptors.

Background Of The Invention

Neuropeptide Y (NPY) is a 36 amino acid peptide that mediates a variety of physiological effects in humans and other mammals. This peptide was first isolated by Tatemoto et al. (*Nature* (1982) 296:659) and subsequently found to be largely conserved across mammalian species. It belongs to a large family of peptides that includes, among others, peptide YY (PYY) and pancreatic peptide (PP). NPY is the most abundant peptide in the mammalian brain, and is also present in sympathetic neurons. In addition, NPY-containing fibers have been found in peripheral tissues, such as around the arteries in the heart, the respiratory tract, the gastrointestinal tract and the genitourinary tract.

Central injection of NPY elicits a multitude of physiological responses, such as stimulation of feeding, increase in fat storage, elevation of blood sugar and insulin, anxiolytic behaviors, reduction in locomotor activity, hormone release, increase in blood pressure, reduction in body temperature and catalepsy. In the cardiovascular system, NPY appears to be involved in the regulation of coronary tone. These effects are selectively mediated by various NPY receptors, which currently include the Y₁, Y₂, Y₃, Y₄, Y₅ and Y₆ subtypes, as well as the hypothetical Y_{1-like} subtype (e.g., Wahlestedt and Reis (1993) *Ann. Rev. Pharmacol. Toxicol.* 33:309; Gehlert and Hipskind (1995) *Curr. Pharm. Design*, 1:295; Michel et al. (1998) *Pharmacol. Rev.* 50:143).

The Y₅ receptor subtype (e.g., U.S. Patent No. 5,602,024) appears to be involved in appetite regulation, including the modulation of food intake and energy expenditure. In addition, studies of seizure-prone mice have suggested that the NPY₅ receptor may have an anti-epileptic activity in the control of limbic seizures. NPY₅-like receptors have also been implicated in attenuation of morphine withdrawal symptoms, enhancement of diuresis and natriuresis, lowering of blood glucose, inhibition of ...

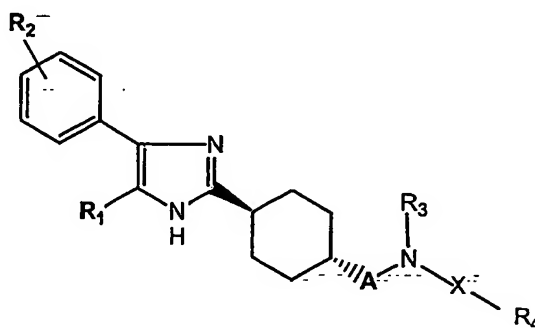
- 2 -

luteinizing hormone secretion, and reduction of acetylcholine release in the ileum. See, for example, Hu et al.(1996) *J. Biol. Chem.*, 271:26315-19; Gerald et al.(1996) *Nature*, 382:168-71; Blomquist et al. (1997) *TINS*, 20: 294-98.

Selective peptide agonists and antagonists have been identified for most of the NPY receptor subtypes. Peptides, however, generally have serious shortcomings for therapeutic use including, poor metabolic stability, low oral bioavailability and poor brain permeability. To date, few non-peptide antagonists have been reported. WO 01/44201 describes certain substituted imidazole NPY receptor antagonists, but additional antagonists with improved properties are needed as therapeutic agents for the treatment of physiological disorders associated with NPY5 receptor activation, such as feeding disorders (e.g., obesity and bulimia), psychiatric disorders, diabetes and cardiovascular diseases (such as hypertension). The present invention fulfills this need, and provides further related advantages.

Summary Of The Invention

The present invention provides NPY5 receptor modulators that inhibit or enhance NPY binding to NPY5 receptor. Such modulators generally comprise a substituted 2-cyclohexyl-4-phenyl-1H-imidazole derivative characterized by the formula:



Formula I

or a pharmaceutically acceptable salt thereof. Within Formula I, R₁ represents (within certain embodiments) hydrogen, C₁-C₈ alkyl, C₂-C₈ alkenyl, C₂-C₈ alkynyl, C₁-C₈ alkoxy, C₁-C₈ alkylthio, halo(C₁-C₈)alkyl, halo(C₁-C₈)alkoxy, hydroxy(C₁-C₈)alkyl, hydroxy(C₁-C₈)alkoxy, mono or di(C₁-C₈)alkyl amino, C₁-C₈ alkanoyl, C₁-C₈ alkoxycarbonyl, C₁-C₈ alkanoyloxy, C₁-C₈ carbonate, C₁-C₈ carbamate, -COOH, -SO₂NH₂, mono or dialkylsulfonamido, -C(O)NH₂ or mono or di(C₁-C₈)alkylcarboxamido; within other embodiments, R₁ is taken together with an R₂

- 3 -

substituent (preferably in an ortho position) to represent a 5- to 7-member carbocyclic or heterocyclic ring that is fused to the phenyl and imidazole rings of formula I. R_1 is preferably hydrogen, C_1 - C_6 alkyl or a group that, taken with R_2 , forms a six-membered ring.

- 5 R_2 of Formula I represents 0 to 5 ring substituents, each substituent independently selected from hydrogen, halogen, hydroxyl, amino, cyano, nitro, C_1 - C_8 alkyl, C_2 - C_8 alkenyl, C_2 - C_8 alkynyl, C_1 - C_6 alkoxy, C_1 - C_8 alkylthio, halo(C_1 - C_8)alkyl, halo(C_1 - C_8)alkoxy, hydroxy(C_1 - C_8)alkyl, hydroxy(C_1 - C_8)alkoxy, mono or di(C_1 - C_8)alkyl amino, C_1 - C_8 alkanoyl, C_1 - C_8 alkanoyloxy, C_1 - C_8 carbonate, C_1 - C_8 carbamate, C_1 - C_8 alkoxycarbonyl, $-COOH$, $-SO_2NH_2$, mono and dialkylsulfonamido, $-C(O)NH_2$,
10 mono and di(C_1 - C_8)alkylcarboxamido, and substituents that (taken together with R_1) form a 5- to 7-member carbocyclic or heterocyclic ring that is fused to the benzene and imidazole rings of Formula I.

- A represents $-C(O)-$ or $-(CH_2)_n-$, wherein n is an integer ranging from 1
15 to 3. Within certain preferred embodiments A is $-C(O)-$ or $-CH_2-$.

- Within Formula I, R_3 represents (i) hydrogen; (ii) C_1 - C_8 alkyl, optionally substituted with from 1 to 6 substituents that are independently selected from halogen, hydroxyl, carbocyclic groups and heterocyclic groups, wherein each carbocyclic or heterocyclic group contains from 3 to 10 ring members; or (iii) a bond to R_4 , forming a
20 heterocyclic group having from 1 to 3 fused or pendant rings, each ring containing from 5 to 10 ring members, wherein each ring is optionally substituted by from 1 to 5 substituents that are independently selected from halogen, cyano, nitro, C_1 - C_8 alkyl, C_2 - C_8 alkenyl, C_2 - C_8 alkynyl, C_1 - C_8 alkoxy, C_1 - C_8 alkylthio, hydroxy, amino, mono or di(C_1 - C_8)alkyl amino, halo(C_1 - C_8)alkyl, halo(C_1 - C_8)alkoxy, hydroxy(C_1 - C_8)alkyl,
25 hydroxy(C_1 - C_8)alkoxy C_2 - C_8 alkanoyl, C_1 - C_8 alkoxycarbonyl, $-COOH$, $-SO_2NH_2$, mono or dialkylsulfonamido, $-C(O)NH_2$ and mono and di(C_1 - C_8)alkylcarboxamido. Within certain preferred embodiments, R_3 is hydrogen, methyl, ethyl or propyl.

X of formula I represents a bond, $-S(O)_2-$, $-C(O)-$ or $-NHC(O)-$.

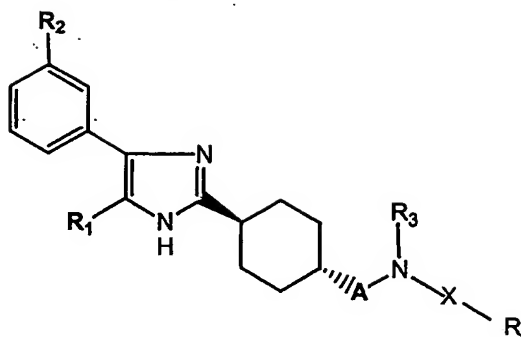
- Within Formula I, R_4 (i) represents hydrogen; (ii) represents C_1 - C_8 alkyl,
30 C_2 - C_8 alkenyl, C_2 - C_8 alkynyl, C_1 - C_8 alkoxy, C_1 - C_8 alkanoyloxy, C_1 - C_8 carbonate, halo(C_1 - C_8)alkyl, halo(C_1 - C_8)alkoxy, hydroxy(C_1 - C_8)alkyl, hydroxy(C_1 - C_8)alkoxy, C_1 - C_8 alkanone or C_1 - C_8 alkyl ether; optionally substituted with from 1 to five substituents that are independently selected from halogen, hydroxyl, carbocyclic groups and

- 4 -

heterocyclic groups, wherein each carbocyclic or heterocyclic group contains from 5 to 10 ring members; or (iii) alone or taken together with R₃, X and the N to which R₃ is bonded, represents a carbocyclic or heterocyclic group having from 1 to 3 fused or pendant rings, each ring containing from 5 to 10 ring members, wherein each ring is optionally substituted by from 1 to 5 substituents that are independently selected from halogen, cyano, nitro, C₁-C₈ alkyl, C₂-C₈ alkenyl, C₂-C₈ alkynyl, C₁-C₈ alkoxy, C₁-C₈ alkanoyloxy, C₁-C₈ carbonate, C₁-C₈ carbamate, C₁-C₈ alkylthio, hydroxy, amino, mono or di(C₁-C₈)alkyl amino, halo(C₁-C₈)alkyl, halo(C₁-C₈)alkoxy, hydroxy(C₁-C₈)alkyl, hydroxy(C₁-C₈)alkoxy C₂-C₈ alkanoyl, C₁-C₈ alkoxycarbonyl, -COOH, -SO₂NH₂, mono or dialkylsulfonamido, -C(O)NH₂ and mono or di(C₁-C₈)alkylcarboxamido. Within certain preferred embodiments, R₄ is (i) an aromatic group selected from phenyl, benzyl, phenoxyl, benzoxyl, phenylethanonyl, pyrimidin-2-yl, tetrahydropyran-2-yl, 2-hydroxy-indan-1-yl, tetrazolyl, pyrimidin-2-yl, pyrimidin-4-yl, pyrimidin-5-yl, pyridin-2-yl, pyridin-3-yl and pyridin-4-yl, wherein the group is optionally substituted by from 1 to 3 substituents independently selected from hydroxyl, halogen and C₁-C₆ alkyl; (ii) a non-aromatic group selected from hydrogen, C₁-C₆ alkyl, C₁-C₆ alkoxy, C₁-C₈ alkanoyloxy, C₁-C₈ carbonate, halo(C₁-C₆)alkyl, halo(C₁-C₆)alkoxy, hydroxy(C₁-C₆)alkyl, hydroxy(C₁-C₆)alkoxy, C₁-C₆ alkanone or C₁-C₆ alkyl ether; or (iii) taken with X, R₃ and the N bonded to R₃ to form a heterocyclic group selected from tetrazolyl, morpholin-4-yl, pyrimidin-2-yl, pyridin-2-yl, pyridin-3-yl and pyridin-4-yl, piperdin-1-yl, pyrrolidin-1-yl, each of which is optionally substituted with from 1 to 3 ring substituents that are independently selected from halogen, C₁-C₆ alkyl, C₁-C₆ alkoxy, hydroxyl, halo(C₁-C₆)alkyl and halo(C₁-C₆)alkoxy.

Within certain preferred embodiments, R₂ is a meta-substituted trifluoromethyl, halogen or cyano. In other words, within such embodiments, the compound further satisfies Formula II, and R₂ is trifluoromethyl, halogen or cyano.

- 5 -

Formula II

- Within certain preferred embodiments, X is SO₂, R₃ is hydrogen and R₄ is a group selected from phenyl, pyridyl and pyrimidyl, wherein the group is optionally substituted with from 1 to 3 substituents independently selected from hydroxyl, halogen and C₁-C₆ alkyl. Within other preferred embodiments, A is -CH₂-, X is CO, R₃ is hydrogen and R₄ is C₁-C₆ alkyl, C₁-C₆ alkoxy, halo(C₁-C₆)alkyl or halo(C₁-C₆)alkoxy. Within still further preferred embodiments, A is CO, X is -NHC(O)-, R₃ is hydrogen and R₄ is C₁-C₆ alkyl. Yet other preferred embodiments include compounds in which A is CO, X is a bond, R₃ is hydrogen or C₁-C₆ alkyl, and R₄ is C₁-C₆ alkyl, halo(C₁-C₆)alkyl, hydroxy(C₁-C₆)alkyl, or an aromatic group selected from phenyl, pyrimidin-2-yl, indan-1-yl, wherein the group is optionally substituted with from 1 to 3 substituents independently selected from hydroxyl, halogen, C₁-C₆ alkyl, hydroxy(C₁-C₆)alkyl and C₁-C₆ alkanone.
- Within certain embodiments, substituted 2-cyclohexyl-4-phenyl-1H-imidazole derivatives provided herein include: (a) 2-Chloro-N-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarbonyl}-benzene sulfonamide; (b) 4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid(2-hydroxy-propyl)-methyl-amide; (c) 4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexane carboxylic acid(2-oxo-2-phenyl-ethyl)-amide; (d) Pyridine-3-sulfonicacid {4-[4-(3-trifluoro-methyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-amide; (e) 4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid(2-hydroxy-2-phenyl-ethyl)-amide; (f) 1-{4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-1H-tetrazole; (g) N-{4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-benzenesulfonamide; (h) 4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid(2-hydroxy-indan-1-yl)-amide; (i) {4-[4-(3-Chloro-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-carbamic acid ethyl

- 6 -

- ester; (j) Cyclopentyl- {4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-amine; (k) N- {4-[4-(4-Bromo-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-2,2,2-trifluoro-acetamide; (l) N- {4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarbonyl}-benzene sulfonamide; (m) Pyrimidin-2-yl- {4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-amine; (n) 4-[4-(3-Bromo-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid N-acetyl-hydrazide; (o) 4,N-Dimethyl-N- {4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarbonyl}-benzene sulfonamide; (p) N- {4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-acetamide; (q) Benzenesulfinic acid {4-[4-(3-chloro-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-amide; (r) 4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid(2-hydroxy-1,1-dimethyl-ethyl)-amide; (s) 4-Hydroxy-N- {4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-butyramide; (t) 4-methyl-N- {4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarbonyl}-benzene sulfonamide; (u) N- {4-[4-(3-Bromo-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-trifluoroacetamide; (v) 4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid (2-hydroxy-propyl)-amide; (w) N- {4-[5-methyl-4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-acetamide; (x) 2,2,2-Trifluoro-N- {4-[4-(4-methoxy-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-acetamide; (y) 4-Chloro-N- {4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarbonyl}-benzene sulfonamide; (z) N-[4-(8-Methoxy-4,5-dihydro-3H-naphtho[1,2-d]imidazol-2-yl)-cyclohexylmethyl]-benzenesulfonamide; (aa) 4-[4-(3-Bromo-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid pyrimidin-2-ylamide; (bb) N-Methyl-N- {4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-benzenesulfonamide; (cc) N- {4-[4-(3-Bromo-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-acetamide; (dd) 5-Methyl-pyridine-2-sulfonic acid {4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarbonyl}-amide; (ee) 4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid (1-hydroxymethyl-2-methyl-propyl)-amide; (ff) {4-[5-Methyl-4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-carbamic acid benzyl ester; (gg) [4-(8-Methoxy-4,5-dihydro-3H-naphtho[1,2-d]imidazol-2-yl)-cyclohexylmethyl]-carbamic acid benzyl ester; (hh) 4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid N-acetyl-hydrazide; (ii) 2,2,2-Trifluoro-N-[4-(4-phenyl-1H-imidazol-2-yl)-cyclohexylmethyl]-acetamide; and (jj) 2,2,2-

- 7 -

Trifluoro-N-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-acetamide.

Within certain embodiments, substituted 2-cyclohexyl-4-phenyl-1H-imidazole derivatives provided herein exhibit a K_i of 1 micromolar or less, 100
5 nanomolar or less, or 10 nanomolar or less in an NPY5 receptor ligand binding assay as provided in Example 11. The ligand (e.g., NPY or PYY) in such assays may be radiolabeled.

Within certain aspects, an NPY5 receptor modulator provided herein comprises a 2-cyclohexyl-4-phenyl-1H-imidazole derivative as described above
10 associated with (*i.e.*, linked to or combined with) a targeting moiety or carrier.

Within further aspects, the present invention provides pharmaceutical compositions comprising a compound or modulator as described above in combination with a physiologically acceptable carrier or excipient. Within certain embodiments, a pharmaceutical composition provided herein may further comprise one or more
15 additional active agents (*i.e.*, drugs). Pharmaceutical compositions provided herein may be formulated, for example, as an injectible fluid, an aerosol, a cream, a gel, a pill, a capsule, a syrup or a transdermal patch.

The present invention further provides, within other aspects, methods for treating a disease or disorder associated with NPY5 receptor activation, comprising
20 administering to a patient in need of such treatment an effective amount of a compound or modulator as described above. Such diseases and disorders include, for example, eating disorders (e.g., obesity and bulimia nervosa), psychiatric disorders, cardiovascular disorders and diabetes. The compound or modulator may be administered orally, or via another means such as intranasally, intravenously or topically. Within
25 certain embodiments, the patient is a human.

Within further aspects, the present invention provides compounds as described above, wherein the compounds are radiolabeled.

Methods are provided, within other aspects, for determining the presence or absence of NPY5 receptor in a sample, comprising the steps of: (a) contacting a
30 sample with an agent comprising a compound as described above under conditions that permit binding of the agent to NPY5 receptor; and (b) detecting a level of agent bound to NPY5 receptor. Within certain embodiments, the agent is a radiolabeled compound, and the step of detection comprises the steps of: (i) separating unbound agent from

- 8 -

bound agent; and (ii) detecting the presence or absence of bound agent in the sample. Detection may be achieved, for example, using autoradiography.

The present invention further provides, within other aspects, methods for modulating binding of NPY to NPY5 receptor. Certain such methods are performed *in vitro*, and comprise contacting NPY5 receptor with a compound or modulator as
5 described above under conditions and in an amount sufficient to detectably modulate NPY binding to NPY5 receptor. Other such methods may be performed *in vivo*, and comprise contacting cells expressing NPY5 receptor with a compound or modulator as described above in an amount sufficient to detectably modulate NPY binding to cells
10 expressing a cloned NPY5 receptor *in vitro*. Modulating of NPY binding may be determined, for example, using a ligand binding assay as provided in Example 11.

Methods are further provided for modulating binding of NPY to NPY5 receptor in a patient, comprising administering to a patient (*i.e.*, a human or non-human animal) a compound or modulator as described above. Patients may include, for
15 example, companion animals such as dogs.

Within certain embodiments of the above methods, the modulation is inhibition and/or the NPY5 receptor is a human NPY5 receptor.

Within further aspects, the present invention provides methods for... modulating the signal-transducing activity of NPY5 receptor, comprising contacting an...
20 NPY5 receptor, either *in vivo* or *in vitro*, with a sufficient amount of an NPY5 receptor modulator, under conditions suitable for binding of NPY to NPY5 receptor.---

Also provided by the present invention are packaged pharmaceutical preparations, comprising: (a) a pharmaceutical composition as described above in a container; and (b) instructions for using the composition to treat a patient suffering from
25 a disease or disorder associated with NPY5 receptor activation. Such disorders include, for example, eating disorders, psychiatric disorders, cardiovascular disorders (such as — hypertension) and diabetes.

These and other aspects of the present invention will become apparent upon reference to the following detailed description. All references disclosed herein are
30 hereby incorporated by reference in their entirety as if each was incorporated... individually.

- 9 -

Detailed Description

As noted above, the present invention provides NPY5 receptor modulators comprising small molecule NPY5 receptor ligands that are 2-cyclohexyl-4-phenyl-1H-imidazole derivatives. Such modulators may be used *in vitro* or *in vivo*, to
5 inhibit or enhance NPY binding to NPY5 receptor in a variety of contexts, discussed in further detail below.

Definitions

Prior to setting forth the invention in detail, it may be helpful to provide definitions of certain terms to be used herein. Compounds of the present invention are
10 generally described using standard nomenclature. For compounds having asymmetric centers, it should be understood that all of the optical isomers and mixtures thereof are encompassed. In addition, compounds with carbon-carbon double bonds may occur in Z- and E- forms; with all isomeric forms of the compounds being included in the present invention. Where a compound exists in various tautomeric forms, the invention is not
15 limited to any one of the specific tautomers, but rather includes all tautomeric forms. Certain compounds are described herein using a general formula that includes variables. Unless otherwise specified, each variable within such a formula is defined independently of other variables.

As used herein, "C₁-C₈ alkyl" refers to straight or branched chain alkyl
20 groups or cycloalkyl groups having 1-8 carbon atoms such as, for example, methyl, ethyl, propyl, isopropyl, n-butyl, sec-butyl, tert-butyl, pentyl, 2-pentyl, isopentyl, neopentyl, hexyl, 2-hexyl, 3-hexyl, 3-methylpentyl, cyclopropyl, cyclopropylmethyl, cyclopentyl, cyclohexyl, cycloheptyl and norbornyl. A C₁-C₈ alkyl substituent may be bonded to an atom within a molecule of interest via any chemically suitable portion of
25 the C₁-C₈ alkyl group. Preferred alkyl groups are C₁-C₆ alkyl groups, especially methyl, ethyl, propyl, butyl, cyclopropyl, cyclopropylmethyl, cyclopentyl and cyclohexyl. Particularly preferred alkyl groups are methyl and ethyl. Similarly, "C₂-C₈ alkenyl" refers to straight or branched chain alkene groups or cycloalkene groups having 2 to 8
carbon atoms. Within an alkenyl group, one or more unsaturated carbon-carbon double
30 bonds are present, and may occur at any stable point along the chain (*e.g.*, ethenyl, allyl and isopropenyl). "C₂-C₈ alkynyl" refers to straight or branched chain alkyne groups having 2 to 8 carbon atoms. Within such a group, one or more unsaturated carbon-carbon triple bonds are present, and may occur at any stable point along the chain (*e.g.*,

- 10 -

ethynyl and propargyl). A "stable point" is bond that, when unsaturated, results in a chemically stable compound (*i.e.*, a compound that can be isolated, characterized and tested for biological activity).

By "C₃-C₁₀ cycloalkyl" is meant alkyl groups having 3-10 carbon atoms forming a mono-, bi-, or polycyclic ring system, such as, for example, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, cycloheptyl and norbornyl. Cycloalkyl groups having 3-8 ring atoms are also encompassed by the term "C₁-C₈ alkyl," but the term "C₃-C₁₀ cycloalkyl" further encompasses 9- and 10-membered rings. Similarly, "cycloalkenyl" or "C₃-C₁₀ cycloalkenyl" refers to hydrocarbon groups having 3-10 carbon atoms forming a mono-, bi-, or polycyclic ring system and containing one or more carbon-carbon double bonds which may occur at any stable point in the ring (*e.g.*, cyclopentenyl, cyclohexenyl or cycloheptenyl).

The term "(cycloalkyl)alkyl" or "(C₃-C₁₀)cycloalkyl(C₁-C₈)alkyl" refers to a straight or branched alkyl substituent having of 1 to 8 carbon atoms that is attached to a mono-, bi-, or polycyclic ring system having 3-10 carbon atoms (*e.g.*, cyclopropylmethyl, cyclobutylmethyl, cyclopentylmethyl, cyclohexylmethyl and cycloheptylmethyl).

"C₂-C₈ alkanoyl" refers to an acyl group with 2 to 8 carbon atoms in a linear, branched or cycloalkyl arrangement, optionally substituted with 1 to 5 substituents independently selected at each occurrence from halogens, cyano, nitro, C₁-C₈ alkyl, C₂-C₈ alkenyl, C₂-C₈ alkynyl, C₁-C₈ alkoxy, C₁-C₈ alkylthio, hydroxy, amino, mono or di(C₁-C₈)alkyl amino, (C₃-C₇)cycloalkyl(C₀-C₃)alkyl, halo(C₁-C₈)alkyl, halo(C₁-C₈)alkoxy, C₁-C₈ alkanoyl, C₁-C₈ alkoxycarbonyl, -COOH, -CONH₂, mono- or di-(C₁-C₈)alkylcarboxamido, -SO₂NH₂, and mono or di(C₁-C₈)alkylsulfonamido.

By "C₁-C₈ alkoxy," in the present invention, is meant an alkyl group of 1 to 8 carbon atoms attached via an oxygen bridge. C₁-C₈ alkoxy groups include, for example, methoxy, ethoxy, propoxy, isopropoxy, n-butoxy, sec-butoxy, tert-butoxy, n-pentoxy, 2-pentoxy, 3-pentoxy, isopentoxy, neopentoxy, hexoxy, 2-hexoxy, 3-hexoxy, and 3-methylpentoxy. C₁-C₆ alkoxy groups are generally preferred, with C₁-C₄ alkoxy groups particularly preferred, especially ethoxy and methoxy. Similarly, "C₁-C₈ alkylthio" refers to an alkyl group of 1 to 8 carbon atoms attached via a sulfur bridge.

The term "C₁-C₈ alkoxycarbonyl" refers to an alkoxy group linked via a carbonyl. In other words, an alkoxycarbonyl group has the general structure --C(=O)-- .

- 11 -

O-alkyl. C₁-C₆ alkyl groups are generally preferred, with C₁-C₄ alkyl groups particularly preferred.

"C₁-C₈ alkanoyloxy," as used herein, refers to an alkanoyl group linked via an oxygen bridge. In other words, an alkanoyloxy group has the general structure –

5 O–C(=O)–alkyl. C₁-C₆ alkyl groups are generally preferred, with C₁-C₄ alkyl groups particularly preferred.

The term "C₁-C₈ carbonate" refers to an alkoxycarbonyl group linked via an oxygen bridge. In other words, a carbonate group has the general structure –O–C(=O)–O–alkyl. C₁-C₆ alkyl groups are generally preferred, with C₁-C₄ alkyl groups particularly preferred.

"C₂-C₈ alkanone" refers to a ketone substituent with 2 to 8 carbon atoms in a linear, branched or cyclic arrangement, optionally substituted with 1 to 5 substituents independently selected at each occurrence from halogens, cyano, nitro, C₁-C₈ alkyl, C₂-C₈ alkenyl, C₂-C₈ alkynyl, C₁-C₈ alkoxy, C₁-C₈ alkylthio, hydroxy, amino, 15 mono or di(C₁-C₈)alkyl amino, (C₃-C₇)cycloalkyl(C₀-C₃)alkyl, halo(C₁-C₈)alkyl, halo(C₁-C₈)alkoxy, C₁-C₈ alkanoyl, C₁-C₈ alkoxycarbonyl, –COOH, –CONH₂, mono- or di-(C₁-C₈)alkylcarboxamido, –SO₂NH₂, and mono or di(C₁-C₈)alkylsulfonamido.

Similarly, "C₂-C₈ alkyl ether" refers to an ether substituent with 2 to 8 carbon atoms, optionally substituted with 1 to 5 substituents independently selected at 20 each occurrence from halogens, cyano, nitro, C₁-C₈ alkyl, C₂-C₈ alkenyl, C₂-C₈ alkynyl, C₁-C₈ alkoxy, C₁-C₈ alkylthio, hydroxy, amino, mono or di(C₁-C₈)alkyl amino, (C₃-C₇)cycloalkyl(C₀-C₃)alkyl, halo(C₁-C₈)alkyl, halo(C₁-C₈)alkoxy, C₁-C₈ alkanoyl, C₁-C₈ alkoxycarbonyl, –COOH, –CONH₂, mono- or di-(C₁-C₈)alkylcarboxamido, –SO₂NH₂, and mono or di(C₁-C₈)alkylsulfonamido. Such a substituent is attached via a carbon 25 atom on either side of the ether linkage.

The term "halogen" includes fluorine, chlorine, bromine and iodine. A "haloalkyl" may be a branched or straight-chain saturated aliphatic hydrocarbon group, substituted with 1 or more halogen atoms. "Halo(C₁-C₈)alkyl" groups have 1 to 8 carbon atoms; "halo(C₁-C₆)alkyl" groups have 1 to 6 carbon atoms. Examples of 30 haloalkyl groups include, but are not limited to, mono-, di- or tri-fluoromethyl; mono-, di- or tri-chloromethyl; mono-, di-, tri-, tetra- or penta-fluoroethyl; and mono-, di-, tri-, tetra- or penta-chloroethyl. Typical haloalkyl groups are trifluoromethyl and difluoromethyl. Preferably not more than 5, and more preferably not more than 3,

- 12 -

haloalkyl groups are present in compounds provided herein. The term "haloalkoxy" refers to a haloalkyl group as defined above attached via an oxygen bridge. "Halo(C₁-C₈)alkoxy" groups have 1 to 8 carbon atoms.

The term "hydroxy(C₁-C₈)alkyl" (or "hydroxy(C₁-C₆)alkyl") refers to
 5 aliphatic group having from 1 to 8 (or 1 to 6) carbon atoms, and further comprising at least one hydroxyl group on the main carbon chain and/or on a side chain. Hydroxy(C₁-C₈)alkyl groups include, for example, 2-hydroxy-1,1-dimethyl-ethyl, 1-hydroxymethyl-2-methyl-propyl and 2-hydroxy-propyl.

The term "C₁-C₈ carbamate," as used herein, refers to a group having the
 10 general structure -N-C(=O)-O-alkyl. C₁-C₆ alkyl groups are generally preferred, with C₁-C₄ alkyl groups particularly preferred.

A "heteroatom," as used herein, is oxygen, sulfur or nitrogen.

A "carbocyclic group" is ring formed entirely by carbon-carbon bonds.

Unless otherwise specified, such a ring may be aromatic or non-aromatic.

15 Representative examples of carbocyclic groups are cycloalkyl groups (e.g., cyclopentane and cyclohexane), as well as aromatic groups such as phenyl, benzyl, naphthyl, phenoxy, benzoyl and phenylethanonyl. Carbon atoms present within a carbocyclic group may, of course, be further bonded to a variety of ring substituents, such as hydrogen, a halogen, cyano, nitro, C₁-C₈ alkyl, C₂-C₈ alkenyl, C₂-C₈ alkynyl,
 20 C₁-C₈ alkoxy, C₁-C₈ alkylthio, hydroxy, amino, mono or di(C₁-C₈)alkyl amino, (C₃-C₇)cycloalkyl(C₀-C₃)alkyl, halo(C₁-C₈)alkyl, halo(C₁-C₈)alkoxy, C₁-C₈ alkanoyl, C₁-C₈ alkoxycarbonyl, -COOH, -CONH₂, mono- or di-(C₁-C₈)alkylcarboxamido, -SO₂NH₂, and mono or di(C₁-C₈)alkylsulfonamido.

A "heterocyclic group" comprises a ring in which at least one ring atom
 25 is a heteroatom (*i.e.*, N, O or S), and the remainder of the ring atoms are carbon. Preferably, a heterocyclic group comprises 1-4 heteroatoms; within certain embodiments 1 or 2 heteroatoms is preferred. A heterocyclic group generally has from 1 to 3 fused or pendant rings, preferably one ring or two fused rings. Typically, each ring contains from 5 to 10 ring members, and may be optionally substituted with from 1
 30 to 5 substituents such as halogen, cyano, nitro, C₁-C₈ alkyl, C₂-C₈ alkenyl, C₂-C₈ alkynyl, C₁-C₈ alkoxy, C₁-C₈ alkylthio, hydroxy, amino, mono or di(C₁-C₈)alkyl amino, halo(C₁-C₈)alkyl, halo(C₁-C₈)alkoxy, hydroxy(C₁-C₈)alkyl, hydroxy(C₁-C₈)alkoxy, C₂-C₈ alkanoyl, C₁-C₈ alkoxycarbonyl, COOH, -SO₂NH₂, mono or dialkylsulfonamido,

- 13 -

-C(O)NH₂ or mono or di(C₁-C₈)alkylcarboxamido. Unless otherwise specified, a heterocyclic group may be aromatic or nonaromatic. As with a carbocyclic group, atoms within a heterocyclic ring may be further linked to a variety of ring substituents.

Examples of heterocyclic groups include, but are not limited to, acridinyl, azocinyl, benzimidazolyl, benzofuranyl, benzo-thio-furanyl, benzothiophenyl, benzoxazolyl, benzothiazolyl, benzotriazolyl, benztetrazolyl, benzisoxazolyl, benzisothiazolyl, benzimidazolyl, carbazolyl, NH-carbazolyl, carbolinyl, chromanyl, chromenyl, cinnolinyl, decahydroquinolinyl, dithiazinyl, dihydrofurotetrahydrofuran, furanyl, furazanyl, imidazolidinyl, imidazolyl, indazolyl, indolenyl, indolinyl, indolizyl, indolyl, isobenzofuranyl, isochromanyl, isoindazolyl, isoindolinyl, isoindolyl, isoquinolinyl, isothiazolyl, isoxazolyl, morpholinyl, naphthyridinyl, octahydroisoquinolinyl, oxadiazolyl, oxazolidinyl, oxazolyl, oxazolidinyl, pyrimidinyl, phenanthridinyl, phenanthrolinyl, phenazinyl, phenothiazinyl, phenoxathiinyl, phenoxazinyl, phthalazinyl, piperazinyl, piperidinyl, pteridinyl, purinyl, pyranyl, pyrazinyl, pyrazolidinyl, pyrazolinyl, pyrazolyl, pyridazinyl, pyridoaxazole, pyridoimidazole, pyridothiazole, pyridinyl, pyridyl, pyrimidinyl, pyrrolidinyl, pyrrolinyl, pyrrolyl, quinazolinyl, quinolinyl, quinoxalinyl, quinuclidinyl, tetrahydrofuranyl, tetrahydroisoquinolinyl, tetrahydroquinolinyl, thiadiazinyl, thiadiazolyl, thianthrenyl, thiazolyl, thienyl, thienothiazolyl, thienooxazolyl, thienoimidazolyl, thiophenyl, triazinyl and xanthenyl. It will be apparent that any such heterocyclic groups may be substituted with one or more substituents as described above.

Preferred heterocyclic groups include, for example, pyrimidinyl (*e.g.*, pyrimidin-2-yl), pyridinyl (pyridin-2-yl, pyridin-3-yl and pyridin-4-yl), morpholinyl (*e.g.*, morpholin-4-yl), piperidinyl (*e.g.*, piperidin-1-yl), pyrrolidinyl (*e.g.*, pyrrolidin-1-yl), tetrazolyl, triazinyl, imidazolyl, oxazolyl, isoxazolyl, indolyl, pyrrolyl, pyrazolyl, quinolinyl, isoquinolinyl, thiazolyl, benzothiadiazolyl, triazolyl, pyrazinyl, furanyl, thienyl, benzothieryl, benzofuranyl, tetrahydropyranyl, indanyl, and substituted derivatives of the foregoing such as methyl-tetrahydropyran-2-yl and 2-hydroxy-indan-1-yl.

A "substituent," as used herein, refers to a molecular moiety that is covalently bonded to an atom within a molecule of interest. For example, a "ring substituent" may be a moiety such as a halogen, alkyl group, haloalkyl group or other

- 14 -

group as discussed herein, that is covalently bonded to an atom (preferably a carbon or nitrogen atom) that is a ring member. The term "substitution" refers to replacing a hydrogen atom in a molecular structure with a substituent as described above, such that the valence on the designated atom is not exceeded, and such that a chemically stable compound (*i.e.*, a compound that can be isolated, characterized, and tested for biological activity) results from the substitution.

The term "NPY receptor" refers to a protein comprising any NPY receptor polypeptide sequence, with mammalian and especially human and monkey sequences generally preferred. "NPY5 receptor" refers to a protein comprising a NPY receptor subtype Y₅ sequence, such as those described within U.S. Patent No. 5,602,024 and herein. An NPY or NPY5 receptor may consist entirely of a naturally-occurring sequence; or may comprise additional components (*e.g.*, N-terminal leader sequence) that do not substantially inhibit the receptor's ability to bind ligand (*i.e.*, at least 50% of the binding affinity of the receptor for NPY and/or PYY is retained). For example, a chimeric NPY5/NPY1 receptor, as described herein, is considered to be an NPY5 receptor. Similarly, truncated NPY receptor sequences, or sequences containing amino acid deletions, substitutes, additions or modifications may be used, provided that NPY receptor binding properties are not substantially diminished (*i.e.*, at least 50% of the endogenous ligand-binding affinity is retained). The binding affinity of a candidate NPY receptor for ligand may be evaluated using a standard binding assay as provided herein (see *also* J. Clin. Invest. (1998) 102:2136).

A "NPY5 receptor modulator," also referred to herein as a "modulator," is a compound that modulates (*i.e.*, increases or decreases) ligand binding to NPY5 receptor. In other words, a modulator may be an NPY5 receptor antagonist or agonist. Modulators comprise a compound that is a 2-cyclohexyl-4-phenyl-1H-imidazole derivative having NPY5 receptor modulating activity. A modulator may consist entirely of such a compound, or may further comprise one or more additional moieties, provided that the modulating activity of the active compound is not substantially diminished (*i.e.*, the ability to increase or decrease ligand binding to NPY5 receptor, as determined using a binding assay provided herein, is not diminished by more than 50%). Such additional moieties include, for example, targeting moieties, other active agents and carriers, any of which may be linked to the active compound via a variety of standard techniques including direct condensation, or by way of bi- or multi-functional linkers.

- 15 -

Alternatively, such additional moieties may be combined with the active compound, without covalent linking. A modulator binds "specifically" to NPY5 receptor if it binds human NPY5 receptor (total binding minus nonspecific binding) with a K_i that is 10-fold, preferably 100-fold, and more preferably 1000-fold, less than the K_i measured for modulator binding to other NPY receptors, such as NPY1. A modulator binds with "high affinity" if the K_i at an NPY receptor is less than 1 micromolar, preferably less than 100 nanomolar or 10 nanomolar. Binding assays for evaluating K_i may be performed, for example, using the human *in vitro* NPY5 binding assay provided herein. Ligand binding to NPY1 receptor may be inhibited within such assays using well known techniques, such as through the use of Thomae compound, as described herein. It will be apparent that either NPY or PYY may be used as the ligand within binding assays.

A "targeting moiety," as used herein is a substance (*e.g.*, a compound or a cell) that increases the local concentration of a modulator in the vicinity of a target site in a patient. There are a wide variety of targeting moieties known in the art, including antibodies and fragments thereof, receptors, ligands and other molecules that bind to cells of, or close to, a target tissue.

A "carrier," "carrier group" or "carrier molecule" is a substance that may be associated with an active compound prior to administration to a patient, generally for the purpose of controlling stability or bioavailability of the compound. Carriers for use within such formulations are generally biocompatible, and may also be biodegradable. Carriers include, for example, monovalent or multivalent molecules such as serum albumin (*e.g.*, human or bovine), egg albumin, peptides, polylysine and polysaccharides such as aminodextran and polyamidoamines. Carriers also include solid support materials such as beads and microparticles comprising, for example, polylactate polyglycolate, poly(lactide-co-glycolide), polyacrylate, latex, starch, cellulose or dextran. A carrier may bear the compounds in a variety of ways, including covalent bonding either directly or via a linker group, noncovalent interaction or admixture.

A moiety is "associated with" an active compound if the moiety is linked to (covalently or noncovalently) or combined with the active compound.

A "linker," as used herein, is any molecule that does not comprise a compound that modulates NPY binding to an NPY5 receptor, and that can be covalently linked to at least two chemical moieties. Linkers may be used to link another moiety to...

- 16 -

a compound that modulates NPY binding to an NPY5 receptor. In general, a linker is bi-functional or multi-functional (*e.g.*, a branched structure). Numerous linkers are known in the art, and may be incorporated into an NPY receptor modulator using any appropriate method, which will be apparent to those of ordinary skill in the art.

5 A "prodrug" is a compound that does not fully satisfy the structural requirements of the compounds provided herein, but is modified *in vivo*, following administration to a patient, to produce an active compound of the present invention. For example, a prodrug may be an acylated derivative of a compound as provided herein. Prodrugs include compounds wherein hydroxy, amine or sulfhydryl groups are bonded
10 to any group that, when administered to a mammalian subject, cleaves to form a free hydroxyl, amino, or sulfhydryl group, respectively. Examples of prodrugs include, but are not limited to, acetate, formate and benzoate derivatives of alcohol and amine functional groups within the compounds provided herein.

A "patient" is any individual treated with a NPY5 receptor modulator as
15 provided herein. Patients include humans, as well as other animals such as companion animals and livestock. Patients may be afflicted with a condition associated with undesirable NPY5 receptor activation, or may be free of such a condition (*i.e.*, treatment may be prophylactic).

NPY5 Receptor Modulators

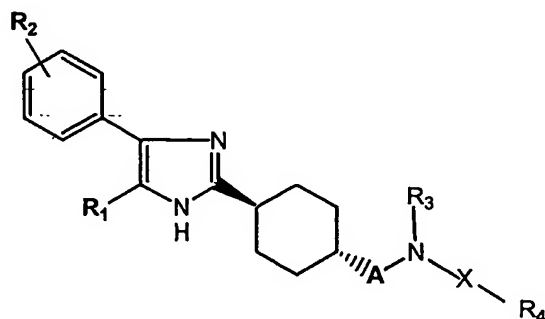
20 As noted above, the present invention provides neuropeptide Y₅ (NPY5) receptor modulators (*i.e.*, agents that detectably modulate both ligand binding to NPY5 and NPY5 receptor-mediated signal transduction). Such modulators may be specific for NPY5 (*i.e.*, do not detectably modulate ligand binding to other NPY receptors), or may also inhibit or enhance ligand binding to one or more additional NPY receptors, such as
25 NPY1. NPY5 receptor modulators may be used to modulate NPY binding to NPY5 *in vivo*, especially in the treatment of feeding disorders (*e.g.*, obesity and bulimia), psychiatric disorders, diabetes and cardiovascular diseases in humans, domesticated companion animals and livestock animals. Modulators may also be used within a variety of *in vitro* assays, such as assays for receptor activity, as probes for detection and
30 localization of NPY5 receptors and as standards in assays of NPY binding and NPY-mediated cellular functions.

The NPY5 receptor modulators provided herein comprise active compounds that are substituted derivatives of 2-cyclohexyl-4-phenyl-1H-imidazole,

- 17 -

which detectably modulate the binding of NPY to NPY5 receptor at nanomolar concentrations, preferably at subnanomolar concentrations. Certain active compounds bind specifically and/or with high affinity to NPY5 receptor. Active compounds may include receptor agonists and antagonists.

- 5 The present invention is based, in part, on the discovery that small molecules having the general formula I (as well as pharmaceutically acceptable salts and prodrugs thereof) modulate NPY binding to NPY5 receptor. Within the context of the present invention, it has been found that the cyclohexyl group in Formula I provides enhanced activity, compared to a heterocyclic ring.

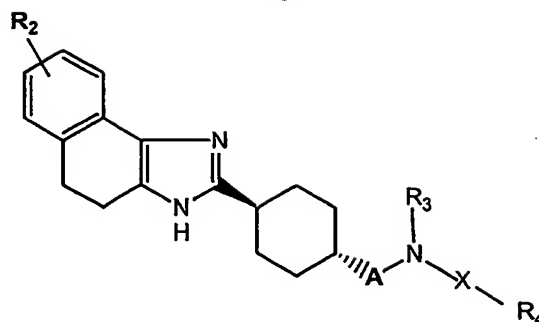


10

Formula I

- Within Formula I, R₁ represents, within certain embodiments, a nonaromatic group such as hydrogen, C₁-C₈ alkyl, C₂-C₈ alkenyl, C₂-C₈ alkynyl, C₁-C₈ alkoxy, C₁-C₈ alkylthio, halo(C₁-C₈)alkyl, halo(C₁-C₈)alkoxy, hydroxy(C₁-C₈)alkyl, hydroxy(C₁-C₈)alkoxy, mono or di(C₁-C₈)alkyl amino, C₁-C₈ alkanoyl, C₁-C₈ alkoxy carbonyl, C₁-C₈ alkanoyloxy, C₁-C₈ carbonate, C₁-C₈ carbamate, -COOH, -SO₂NH₂, mono or dialkylsulfonamido, -C(O)NH₂ or mono or di(C₁-C₈) alkylcarboxamido. Alternatively, within other embodiments, R₁ is taken together with R₂, to form a carbocyclic or heterocyclic ring that is fused to the phenyl and imidazole rings of Formula I. The phrase "R₁ taken together with R₂," as used herein, is intended to refer to a covalent attachment between R₁ and R₂. It will be apparent to those of ordinary skill in the art that a ring formed by such an attachment also includes two carbon atoms that are members of the phenyl group, and two carbon atoms that are members of the imidazole ring of Formula I. One such representative structure is shown in Formula III. A ring generated by R₁ taken together with R₂ typically has from 5 to 7 ring members. Preferred R₁ groups include hydrogen, C₁-C₆ alkyl and groups that, when taken together with R₂, form a six-membered ring.

- 18 -

Formula III

R_2 , within Formula I, represents 0 to 5 ring substituents (*i.e.*, 0 or 1 substituent at each carbon atom of the benzene ring), preferably 0 to 3 substituents, and more preferably 0 or 1 substituent. Each optional, independently selected substituent may be a nonaromatic group such as hydrogen, halogen, hydroxyl, amino, cyano, nitro, C_1 - C_8 alkyl, C_2 - C_8 alkenyl, C_2 - C_8 alkynyl, C_1 - C_8 alkoxy, C_1 - C_8 alkanoyloxy, C_1 - C_8 carbonate, C_1 - C_8 carbamate, C_1 - C_8 alkylthio, halo(C_1 - C_8)alkyl, halo(C_1 - C_8)alkoxy, hydroxy(C_1 - C_8)alkyl, hydroxy(C_1 - C_8)alkoxy, mono or di(C_1 - C_8)alkyl amino, C_1 - C_8 alkanoyl, C_1 - C_8 alkoxycarbonyl, $-COOH$, $-SO_2NH_2$, mono and dialkylsulfonamido, $-C(O)NH_2$, mono and di(C_1 - C_8)alkylcarboxamido. Alternatively, an R_2 substituent may be taken together with R_1 to form a carbocyclic or heterocyclic ring that is fused to the phenyl and imidazole rings of Formula I, such as that shown in Formula II. As noted above, a ring so generated typically has from 5 to 7 ring members. Preferred R_2 groups include meta-substituted trifluoromethyl, halogen or cyano groups, as well as groups that, when taken together with R_1 , form a six-membered ring. It will be apparent to those of ordinary skill in the art that one R_2 group may form a fused ring with R_1 , while one or more other R_2 groups may be located elsewhere on the phenyl ring. It will be further apparent that combinations of R_2 substituents are permissible only if such combinations result in stable compounds. A stable compound or stable structure is meant to imply a compound that is sufficiently robust to survive isolation, characterization and testing for biological activity.

A, in Formula I, represents $-C(O)-$ or $-(CH_2)_n-$, wherein n is an integer ranging from 1 to 3, and preferably is $-C(O)-$ or $-CH_2-$.

R_3 , within certain embodiments, represents hydrogen or C_1 - C_8 alkyl. Such an alkyl group may, but need not, be substituted with from 1 to 8 substituents that are independently selected from halogen, hydroxyl, carbocyclic groups and heterocyclic

- 19 -

groups, wherein each carbocyclic or heterocyclic group contains from 3 to 10 ring members. Within other embodiments, R₃ is a bond to R₄, forming a heterocyclic group that comprises R₃, the nitrogen, X and R₄. Such a heterocyclic group generally contains from 1 to 3 fused or pendant rings, preferably a single ring or two fused rings, and each ring generally contains from 5 to 10 ring members. Each ring within such a heterocyclic group is optionally substituted with from 1 to 5 substituents that are independently selected from halogen, cyano, nitro, C₁-C₈ alkyl, C₂-C₈ alkenyl, C₂-C₈ alkynyl, C₁-C₈ alkoxy, C₁-C₈ alkanoyloxy, C₁-C₈ carbonate, C₁-C₈ carbamate, C₁-C₈ alkylthio, hydroxy, amino, mono or di(C₁-C₈)alkyl amino, halo(C₁-C₈)alkyl, halo(C₁-C₈)alkoxy, hydroxy(C₁-C₈)alkyl, hydroxy(C₁-C₈)alkoxy C₁-C₈ alkanoyl, C₁-C₈ alkoxycarbonyl, -COOH, -SO₂NH₂, mono or dialkylsulfonamido, -C(O)NH₂ and mono and di(C₁-C₈)alkylcarboxamido. R₃ is preferably hydrogen, methyl, ethyl or propyl.

X, within Formula I, represents a bond, -S(O)₂-, -C(O)- or -NHC(O)-.

If X is a bond, then the nitrogen is directly (covalently) bonded to R₄.

R₄ represents, within certain embodiments, represents hydrogen, C₁-C₈ alkyl, C₂-C₈ alkenyl, C₂-C₈ alkynyl, C₁-C₈ alkoxy, C₁-C₈ alkanoyloxy, C₁-C₈ carbonate, halo(C₁-C₈)alkyl, halo(C₁-C₈)alkoxy, hydroxy(C₁-C₈)alkyl hydroxy(C₁-C₈)alkoxy, C₁-C₈ alkanone or C₁-C₈ alkyl ether. Any such group (except hydrogen) is optionally substituted with from 1 to 8 substituents that are independently selected from halogen, hydroxyl, carbocyclic groups and heterocyclic groups. Any such carbocyclic or heterocyclic group contains from 5 to 10 ring members, preferably 5 to 7 ring members. Within other embodiments, R₄ represents a carbocyclic or heterocyclic group that contains from 1 to 3 fused or pendant rings, preferably a single ring or two fused rings, and each ring generally contains from 5 to 10 ring members. Each ring within such a carbocyclic or heterocyclic group is optionally substituted with from 1 to 5 substituents that are independently selected from halogen, cyano, nitro, C₁-C₈ alkyl, C₂-C₈ alkenyl, C₂-C₈ alkynyl, C₁-C₈ alkoxy, C₁-C₈ alkanoyloxy, C₁-C₈ carbonate, C₁-C₈ carbamate, C₁-C₈ alkylthio, hydroxy, amino, mono or di(C₁-C₈)alkyl amino, halo(C₁-C₈)alkyl, halo(C₁-C₈)alkoxy, hydroxy(C₁-C₈)alkyl, hydroxy(C₁-C₈)alkoxy C₁-C₈ alkanoyl, C₁-C₈ alkoxycarbonyl, COOH, -SO₂NH₂, mono or dialkylsulfonamido, -C(O)NH₂ and mono or di(C₁-C₈)alkylcarboxamido. Within still further embodiments, R₄ is taken together with R₃ and X to form a heterocyclic group that comprises R₃, the nitrogen, X and R₄. The phrase "taken together with R₃ and X" is intended to refer to embodiments in which

- 20 -

R₄ is covalently bonded to R₃, forming at least one ring that comprises R₃, the nitrogen, X and R₄. The entire heterocyclic group generally contains from 1 to 3 fused or pendant rings, preferably a single ring or two fused rings, and each ring generally contains from 5 to 10 ring members. Each ring within such a heterocyclic group may, but need not, be substituted by from 1 to 5 substituents that are independently selected from halogen, cyano, nitro, C₁-C₈ alkyl, C₂-C₈ alkenyl, C₂-C₈ alkynyl, C₁-C₈ alkoxy, C₁-C₈ alkanoyloxy, C₁-C₈ carbonate, C₁-C₈ carbamate, C₁-C₈ alkylthio, hydroxy, amino, mono or di(C₁-C₈)alkyl amino, halo(C₁-C₈)alkyl, halo(C₁-C₈)alkoxy, hydroxy(C₁-C₈)alkyl, hydroxy(C₁-C₈)alkoxy C₁-C₈ alkanoyl, C₁-C₈ alkoxycarbonyl, -COOH, -SO₂NH₂, mono or dialkylsulfonamido, -C(O)NH₂ and mono or di(C₁-C₈)alkylcarboxamido.

Preferred R₄ groups include aromatic groups such as phenyl, benzyl, phenoxyl, benzoxyl, phenylethanonyl, pyrimidin-2-yl, pyrimidin-4-yl, pyrimidin-5-yl, tetrahydropyran-2-yl, 2-hydroxy-indan-1-yl, tetrazolyl, pyrimidin-2-yl, pyridin-2-yl, pyridin-3-yl and pyridin-4-yl. Any such groups may, but need not, be substituted by from 1 to 3 substituents independently selected from hydroxyl, halogen and C₁-C₆ alkyl. Other preferred R₄ groups are the non-aromatic groups hydrogen, C₁-C₆ alkyl, C₁-C₆ alkoxy, halo(C₁-C₆)alkyl, halo(C₁-C₆)alkoxy, hydroxy(C₁-C₆)alkyl, hydroxy(C₁-C₆)alkoxy, C₁-C₆ alkanone and C₁-C₆ alkyl ether. Still further preferred R₄ groups are those in which R₄ is taken with X, R₃ and the N bonded to R₃ to form a heterocyclic group such as tetrazolyl, morpholin-4-yl, pyrimidin-2-yl, pyridin-2-yl, pyridin-3-yl and pyridin-4-yl, piperdin-1-yl, pyrrolidin-1-yl and derivatives of the foregoing in which from 1 to 3 ring members are substituted with a substituent independently selected from halogen, C₁-C₆ alkyl, C₁-C₆ alkoxy, hydroxyl, halo(C₁-C₆)alkyl and halo(C₁-C₆)alkoxy.

Certain representative compounds provided herein satisfy one of the following criteria:

- (i) X is SO₂, R₃ is hydrogen and R₄ is a group selected from phenyl, pyridyl and pyrimidyl, wherein the group is optionally substituted by from 1 to 3 substituents independently selected from hydroxyl, halogen and C₁-C₆ alkyl;
- (ii) A is -CH₂-, X is -C(O)-, R₃ is hydrogen and R₄ is C₁-C₆ alkyl, C₁-C₆ alkoxy, halo(C₁-C₆)alkyl or halo(C₁-C₆)alkoxy;
- (iii) A is -C(O)-, X is -NHC(O)-, R₃ is hydrogen and R₄ is C₁-C₆ alkyl; or

- 21 -

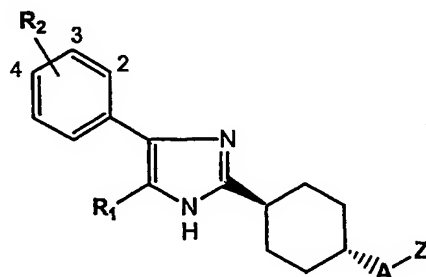
- (iv) A is $-C(O)-$, X is a bond, R_3 is hydrogen or C_1-C_6 alkyl, and R_4 is C_1-C_6 alkyl, halo(C_1-C_6)alkyl, hydroxy(C_1-C_6)alkyl, or an aromatic group selected from phenyl, pyrimidin-2-yl, indan-1-yl, wherein the group is optionally substituted by from 1 to 3 substituents independently selected from hydroxyl, halogen, C_1-C_6 alkyl, hydroxy(C_1-C_6)alkyl and C_1-C_6 alkanone.

Representative compounds provided herein include, but are not limited to, (a) 2-Chloro-N-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarbonyl}-benzene sulfonamide; (b) 4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid(2-hydroxy-propyl)-methyl-amide; (c) 4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexane carboxylic acid(2-oxo-2-phenyl-ethyl)-amide; (d) Pyridine-3-sulfonic acid{4-[4-(3-trifluoro-methyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-amide; (e) 4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid(2-hydroxy-2-phenyl-ethyl)-amide; (f) 1-{4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-1H-tetrazole; (g) N-{4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-benzenesulfonamide; (h) 4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid(2-hydroxy-indan-1-yl)-amide; (i) {4-[4-(3-Chloro-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-carbamic acid ethyl ester; (j) Cyclopentyl-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-amine; (k) N-{4-[4-(4-Bromo-phenyl)-1H-imidazol-2-yl]-cyclohexyl-methyl}-2,2,2-trifluoro-acetamide; (l) N-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarbonyl}-benzene sulfonamide; (m) Pyrimidin-2-yl-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-amine; (n) 4-[4-(3-Bromo-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid N-acetyl-hydrazide; (o) 4,N-Dimethyl-N-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarbonyl}-benzene sulfonamide; (p) N-{4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-acetamide; (q) Benzenesulfinic acid {4-[4-(3-chloro-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-amide; (r) 4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid(2-hydroxy-1,1-dimethyl-ethyl)-amide; (s) 4-Hydroxy-N-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-butyramide; (t) 4-methyl-N-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarbonyl}-benzene sulfonamide; (u) N-{4-[4-(3-Bromo-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-trifluoroacetamide; (v) 4-[4-(3-Trifluoromethyl-

- 22 -

phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid (2-hydroxy-propyl)-amide; (w) N-{4-[5-methyl-4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-acetamide; (x) 2,2,2-Trifluoro-N-{4-[4-(4-methoxy-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-acetamide; (y) 4-Chloro-N-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarbonyl}-benzene sulfonamide; (z) N-[4-(8-Methoxy-4,5-dihydro-3H-naphtho[1,2-d]imidazol-2-yl)-cyclohexylmethyl]-benzenesulfonamide; (aa) 4-[4-(3-Bromo-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid pyrimidin-2-ylamide; (bb) N-Methyl-N-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-benzenesulfonamide; (cc) N-{4-[4-(3-Bromo-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-acetamide; (dd) 5-Methyl-pyridine-2-sulfonic acid {4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarbonyl}-amide; (ee) 4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid (1-hydroxymethyl-2-methyl-propyl)-amide; (ff) {4-[5-Methyl-4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-carbamic acid benzyl ester; (gg) [4-(8-Methoxy-4,5-dihydro-3H-naphtho[1,2-d]imidazol-2-yl)-cyclohexylmethyl]-carbamic acid benzyl ester; (hh) 4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid N-acetyl-hydrazide; (ii) 2,2,2-Trifluoro-N-[4-(4-phenyl-1H-imidazol-2-yl)-cyclohexylmethyl]-acetamide; and (jj) 2,2,2-Trifluoro-N-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-acetamide.

Certain representative compounds encompassed by Formula I are illustrated in Table I. Within Table I, reference is made to Formula IV, and the following abbreviations are used: Ph is phenyl, Me is methyl, Et is ethyl, nPr or Pr is n-propyl, iPr is isopropyl, tBu is tert-butyl, cPent is cyclopentyl, cHex is cyclohexyl.



Formula IV

- 23 -

Table I

Compound	A	R ₁	R ₂	Z
1	CH ₂	H	3-Cl	NH-SO ₂ Ph
2	CH ₂	H	3-Cl	NH-CO ₂ CH ₂ Ph
3	CH ₂	H	3-Cl	NH-CO ₂ CH ₂ CH ₃
4	CH ₂	H	H	NH-SO ₂ CH ₃
5	CH ₂	H	3-CF ₃	-NH ₂
6	CH ₂	H	3-CF ₃	NH-SO ₂ Ph
7	CH ₂	H	3-CF ₃	N(CH ₃)-SO ₂ Ph
8	CH ₂	H	3-CF ₃	NHC=O(CH ₃)
9	CH ₂	H	3-CF ₃	NHCH ₂ CH ₃
10	CH ₂	H	3-CF ₃	NH-cyclopentyl
11	CH ₂	H	3-CF ₃	N-(CH ₂ CH ₂ CH ₃) ₂
12	CH ₂	H	3-Br	NHC=O(CF ₃)
13	CH ₂	H	3-Br	NHC=O(CH ₃)
14	CH ₂	H	CN	NHC=O(CH ₃)
15	CH ₂	H	3-CF ₃	NH-tetrahydropyran-4-yl
16	CH ₂	H	3-CF ₃	NHC=O(CH ₂) ₃ OH
17	CH ₂	H	3-CF ₃	NHC=OCH ₂ CH(OH)CH ₃
18	CH ₂	CH ₃	3-CF ₃	NH-CO ₂ CH ₂ Ph
19	CH ₂	CH ₃	3-CF ₃	NH-cyclopentyl
20	CH ₂	CH ₃	3-CF ₃	NHC=O(CH ₃)
21	CH ₂	H	H	NHC=O(CF ₃)
22	CH ₂	H	4-OCH ₃	NHC=O(CF ₃)
23	CH ₂	H	3-CF ₃	NHC=O(CF ₃)
24	CH ₂	H	H	NHC=O(CH ₃)
25	CH ₂	H	4-Br	NHC=O(CF ₃)
26	CH ₂	H	4-Br	NHC=O(CH ₃)
27	CH ₂	H	3-CF ₃	NH-pyrimidin-2-yl
28	CH ₂	H	H	NH-CH ₂ CF ₃
29	CH ₂	H	4-Br	-NH ₂
30	CH ₂	H	4-Br	NHCH ₂ (tetrahydropyran-2-yl)
31	CH ₂	H	4-Br	morpholin-4-yl
32	CH ₂	H	3-CF ₃	NH-SO ₂ -(pyridin-3-yl)
33	CH ₂	H	H	NHC=O(pyridin-3-yl)
34	CH ₂	H	H	NH-SO ₂ -(pyridin-3-yl)
35	CH ₂	H	3-Cl	NHC=O(pyridin-3-yl)
36	CH ₂	H	3-Cl	NH-SO ₂ -(pyridin-3-yl)
37	C=O	H	3-CF ₃	NHNHC=O(CH ₃)
38	C=O	H	3-CF ₃	NHSO ₂ -(4-CH ₃ -phenyl)
39	C=O	H	3-Br	NHNHC=O(CH ₃)
40	C=O	H	3-Br	NH(CH ₂) ₂ CH ₃
41	C=O	H	3-Br	NH-pyrimidin-2-yl
42	C=O	H	3-Br	N-(CH ₂ CH ₂ CH ₃) ₂
43	C=O	H	3-Br	NHCH ₂ CH(OH)CH ₃
44	C=O	H	3-Br	NHCH ₂ C=O(CH ₃)
45	C=O	H	3-CF ₃	NHSO ₂ -(2-Cl-phenyl)

- 24 -

Compound	A	R ₁	R ₂	Z
46	C=O	H	3-CF ₃	NHSO ₂ -(3-Cl-phenyl)
47	C=O	H	3-CF ₃	NHSO ₂ -(4-Cl-phenyl)
48	C=O	H	3-CF ₃	NHSO ₂ -Ph
49	C=O	H	3-CF ₃	NHSO ₂ -(5-CH ₃ -pyridin-2-yl)
50	C=O	H	3-CF ₃	NHSO ₂ CH ₂ Ph
51	C=O	H	3-CF ₃	NHSO ₂ -(2-CH ₃ -phenyl)
52	C=O	H	3-CF ₃	(R)-NHCH ₂ CH(OH)CH ₃
53	C=O	H	3-CF ₃	(S)-NHCH ₂ CH(OH)CH ₃
54	C=O	H	3-CF ₃	NH-SO ₂ (CH ₂) ₃ CH ₃
55	C=O	H	3-CF ₃	NH-SO ₂ (CH ₂) ₂ CH ₃
56	C=O	H	3-CF ₃	NH-SO ₂ CH ₂ CH ₃
57	C=O	H	3-CF ₃	NH-SO ₂ CH ₂ CF ₃
58	C=O	H	3-CF ₃	NCH ₃ -SO ₂ -(4-CH ₃ -phenyl)
59	C=O	H	3-CF ₃	NH(CH ₂) ₂ OH
60	C=O	H	3-CF ₃	NHC(CH ₃) ₂ CH ₂ OH
61	C=O	H	3-CF ₃	NHCH(CH ₃)CH ₂ OH
62	C=O	H	3-CF ₃	NH(CH ₂) ₃ OH
63	C=O	H	3-CF ₃	NHCH ₂ CH(OH)Ph
64	C=O	H	3-CF ₃	NH(CH ₂) ₂ CH(OH)CH ₃
65	C=O	H	3-CF ₃	NHCH(CH ₂ OH)CH(CH ₃) ₂
66	C=O	H	3-CF ₃	NHCH ₂ CH(OH)CH ₂ CH ₃
67	C=O	H	3-CF ₃	NH-1-(CH ₂ OH)cyclopent-1-yl
68	C=O	H	3-CF ₃	NH(1-OH-cyclohexyl)-methyl
69	C=O	H	3-CF ₃	NH-(2-OH-cyclohex-1-yl)
70	C=O	H	3-CF ₃	NH(CH ₂) ₂ OCH ₃
71	C=O	H	3-CF ₃	NH(CH ₂) ₂ NH ₂
72	C=O	H	3-CF ₃	NH(CH ₂) ₂ N(CH ₃) ₂
73	C=O	H	3-CF ₃	morpholin-4-yl
74	C=O	H	3-CF ₃	NH-cyclohexyl
75	C=O	H	3-CF ₃	NCH ₃ -CH ₂ CH(OH)CH ₃
76	C=O	H	3-CF ₃	(S)-NHCH ₂ CH(OH)Ph
77	C=O	H	3-CF ₃	(R)-NHCH ₂ CH(OH)Ph
78	C=O	H	3-CF ₃	3-(OH)-piperidin-1-yl
79	C=O	H	3-CF ₃	3-(OH)-pyrrolidin-1-yl
80	C=O	H	3-CF ₃	(1R,2S)NH(2-OH-indan-1-yl)
81	C=O	H	3-CF ₃	(1S,2R)NH(2-OH-indan-1-yl)
82	C=O	H	3-CF ₃	4-(OH)-piperidin-1-yl
83	C=O	H	3-CF ₃	NH-CH ₂ CH(OH)C(CH ₃) ₃
84	C=O	H	3-CF ₃	(S)-NH-CH(CH ₂ OH)CH(CH ₃) ₂
85	C=O	H	3-CF ₃	(R)-NH-CH(CH ₂ OH)CH(CH ₃) ₂
86	C=O	H	3-CF ₃	NH-CH ₂ CH(OH)CF ₃

It will be apparent to those of ordinary skill in the art that Table I provides only representative examples of compounds provided herein, and is not intended to limit the scope of the present invention. Further, as noted above, all

- 25 -

compounds of the present invention may be present as a free base or as a pharmaceutically acceptable acid addition salt.

Substituted derivatives of 2-cyclohexyl-4-phenyl-1H-imidazole provided herein detectably alter (modulate) NPY binding to NPY5 receptor, as determined using standard *in vitro* NPY5 receptor ligand binding assays and/or signal transduction assays. References herein to an "NPY5 receptor ligand binding assay" are intended to refer to the protocol provided in Example 11. Briefly, a competition assay may be performed in which an NPY5 receptor preparation is incubated with labeled (*e.g.*, ^{125}I) NPY and unlabeled test compound. Within the assays provided herein, the NPY5 receptor used is preferably a mammalian NPY5 receptor, more preferably a human or monkey NPY5 receptor. The receptor may be recombinantly expressed or naturally expressed, and may comprise a native sequence or a modified sequence (*e.g.*, truncated and/or fused to a non-native N-terminal sequence). The NPY5 receptor preparation may be, for example, a membrane preparation from Sf9 cells or Bowes Melanoma cells that recombinantly express human NPY5 receptor or a human chimeric NPY5/NPY1 receptor.

Incubation with a compound that detectably modulates NPY binding to NPY5 receptor will result in a decrease or increase in the amount of label bound to the NPY5 receptor preparation, relative to the amount of label bound in the absence of the compound. Preferably, such a compound will exhibit a K_i at an NPY5 receptor of less than 1 micromolar, more preferably less than 500 nM, 100 nM, 20 nM or 10 nM, within an assay performed as described in Example 11. Generally preferred compounds are NPY5 receptor antagonists, and decrease NPY5 receptor activity (as measured by calcium mobilization, as described in Example 12) by at least 20%, preferably by at least 50%, and most preferably by at least 80%. For certain uses, preferred compounds also decrease food intake and weight gain in one or more animal models, such as food deprivation models (as described, for example, in published PCT application PCT/US00/26887) and the bovine pancreatic polypeptide antagonism model, as described in Example 13.

If desired, compounds provided herein may be evaluated for certain pharmacological properties including, but not limited to, oral bioavailability, serum protein binding and *in vitro* and *in vivo* half-life. In addition, penetration of the blood brain barrier may be desirable for compounds used to treat CNS disorders, while low brain levels of compounds used to treat peripheral disorders may be preferred. Routine

- 26 -

assays that are well known in the art may be used to assess these properties, and identify superior compounds for a particular use. For example, assays used to predict bioavailability include transport across human intestinal cell monolayers, including Caco-2 cell monolayers. Penetration of the blood brain barrier of a compound in humans may be predicted from the brain levels of the compound in laboratory animals given the compound intravenously. Serum protein binding may be predicted from albumin binding assays, as described, for example, in a review by Oravcová et al. (Journal of Chromatography B (1996) 677:1-27). Compound half-life is inversely proportional to the frequency of dosage of a compound. *In vitro* half-lives of compounds may be predicted from assays of microsomal half-life as described by Kuhnz and Gieschen (Drug Metabolism and Disposition (1998) 26:1120-1127). In view of the present disclosure, a person of ordinary skill in the art could use such routine techniques to select a compound that displays optimal properties for a particular purpose.

As noted above, NPY5 receptor modulators provided herein may comprise, in addition to an active compound of formula I, one or more additional associated moieties. Such moieties may be linked directly (*i.e.*, via a bond) or by way of a linker, may be noncovalently linked or may be combined with the compound. Such additional moieties may be used, for example, to facilitate delivery, targeting or detection of the compound. For example, compounds provided herein may sufficiently target a desired site *in vivo*; however, it may be beneficial for certain applications to include an additional targeting moiety to facilitate targeting to one or more specific tissues. Preferred targeting moieties include those that target to brain regions associated with NPY5 activity.

For certain embodiments, it may be beneficial to also, or alternatively, associate a drug with a modulator. As used herein, the term "drug" refers to any bioactive agent intended for administration to a mammal to prevent or treat a disease or other undesirable condition. Drugs include hormones, growth factors, proteins, peptides and other compounds. For example, modulators for treatment of eating disorders, particularly obesity and bulimia nervosa, may comprise an agent such as sibutramine, dextenfluramine, leptin, a growth hormone secretagogue, a melanocortin agonist, a beta-3 agonist, a 5HT-2 agonist, an orexin antagonist, a melanin concentrating hormone antagonist, a galanin antagonist, a CCK agonist, a GLP-1 agonist, a corticotropin-

- 27 -

releasing hormone agonist or a NPY₁ antagonist. Moieties that facilitate detection include radionuclides, luminescent groups, fluorescent groups and enzymes, all of which may be associated with a compound via standard methods.

For detection purposes, as discussed in more detail below, compounds provided herein may be isotopically-labeled or radiolabeled. Such compounds are identical to those recited in Formula I, but for the fact that one or more atoms are replaced by an atom having an atomic mass or mass number different from the atomic mass or mass number usually found in nature. Examples of isotopes that can be incorporated into compounds provided herein include isotopes of hydrogen, carbon, nitrogen, oxygen, phosphorous, fluorine and chlorine, such as ²H, ³H, ¹¹C, ¹³C, ¹⁴C, ¹⁵N, ¹⁸O, ¹⁷O, ³¹P, ³²P, ³⁵S, ¹⁸F and ³⁶Cl. In addition, substitution with heavy isotopes such as deuterium (*i.e.*, ²H) can afford certain therapeutic advantages resulting from greater metabolic stability, for example increased in vivo half-life or reduced dosage requirements and, hence, may be preferred in some circumstances.

Other moieties that may be associated with an active compound include carriers. Such substances may modulate bioavailability or stability of the compound. Representative carriers include, for example, molecules such as albumin, polylysine, polyamidoamines, peptides, proteins, polystyrene, polyacrylamide, lipids, ceramide and biotin, solid support materials such as beads and microparticles comprising, for example, polylactate polyglycolate, poly(lactide-co-glycolide), polyacrylate, latex, starch, cellulose or dextran.

Preparation of NPY₅ Receptor Modulators

Substituted derivatives of 2-cyclohexyl-4-phenyl-1H-imidazole may generally be prepared using standard synthetic methods, which are well known to those of ordinary skill in the art of organic synthesis. Representative methods are described below, and within Examples 1-9. Such methods may be combined with other known synthetic methods and variations thereon (*e.g.*, modification of starting materials) that will be apparent to those of ordinary skill in the art, to generate all compounds provided herein.

By way of example, a synthetic route similar to those shown in any one of Schemes 1-3 (below) may be used. Within these Schemes, "coupling" refers to a suitable coupling reagent such as, but not limited to BOP or EDCI. These coupling reactions can be carried out at ambient or elevated temperatures using various solvents

- 28 -

including, but not limited to, methylene chloride, DMF and THF. Coupling reactions can be used to prepare the compounds encompassed by general structures 1-C (Scheme 1) and 3-D (Scheme 3). In Scheme 1, following coupling, compound 1-C may be converted directly to compound 1-E (when R₁ is H). Alternatively, to generate
5 compounds with R₁ groups that are not H, synthesis may proceed via compound 1-D.

In Scheme 2, "deprotection" refers to the process of removal of benzyl carbamate group. This deprotection can be carried out in a number of ways well known to those skilled in the art of organic synthesis including, but not limited to, catalytic hydrogenation, acid hydrolysis and base hydrolysis. Deprotection can be used to
10 prepare the compounds encompassed in general structure 2-B.

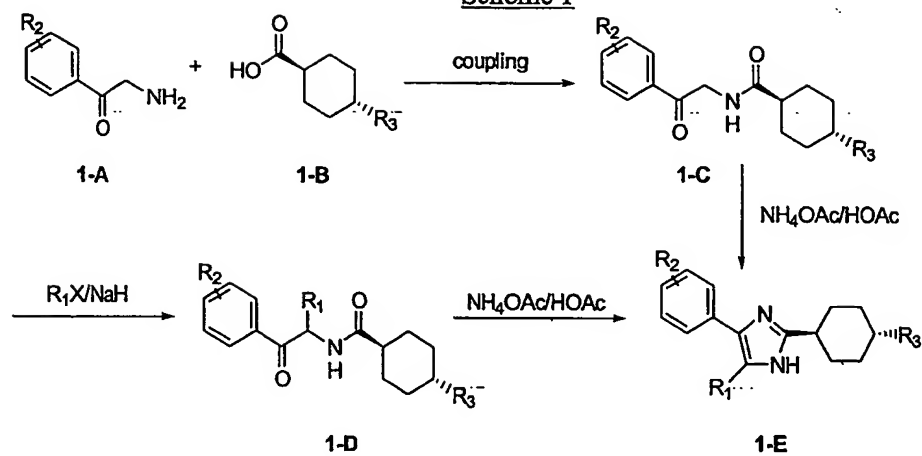
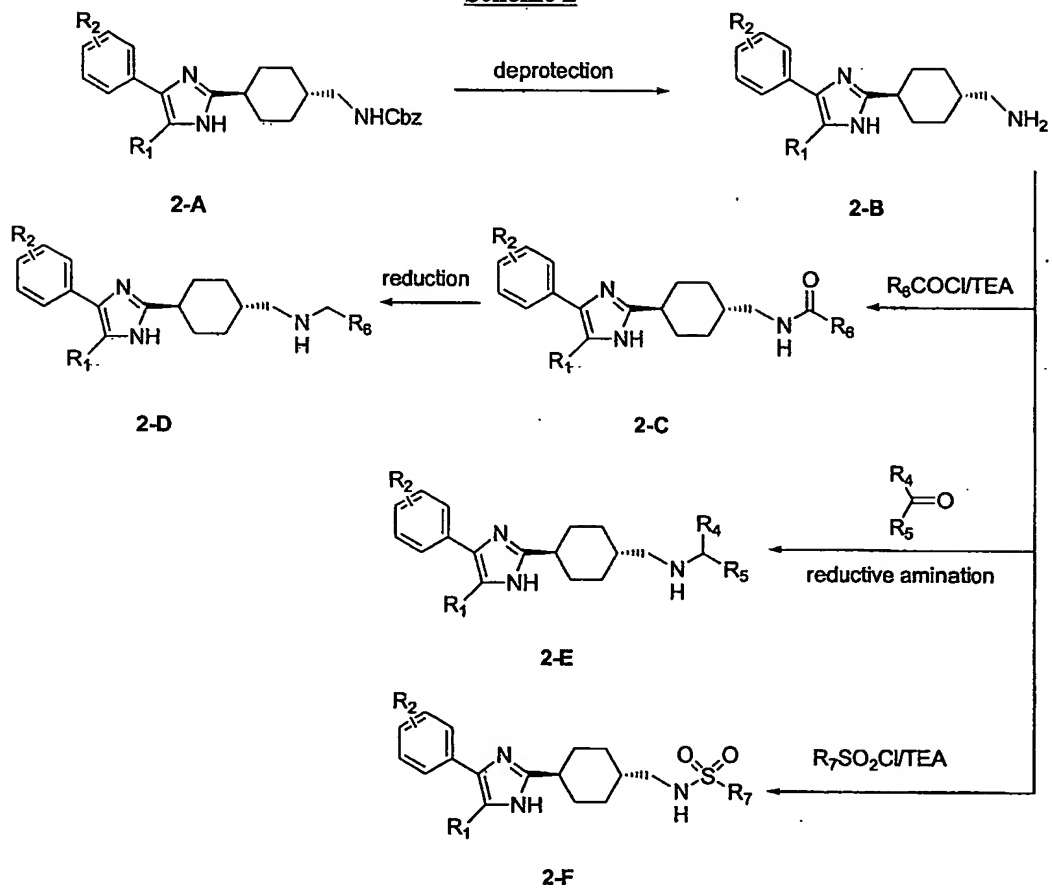
In Scheme 2, "reduction" refers to the process of synthetic transformation wherein an amide moiety is converted to an amine. Such a transformation can be used to prepare compounds of general structure 2-D. Suitable reductive reagents include, but
are not limited to, BH₃, LiAlH₄ and LiBH₃CN.

15 In Scheme 2, "reductive amination" refers to the process of synthetic transformation wherein a primary amine is converted to a secondary amine by reaction with a suitable aldehyde or ketone. Such a transformation can be carried out in a number of ways well known to those skilled in the art of organic synthesis. Suitable reducing reagents include, but are not limited to, BH₃, NaBH₄, NaBH₃CN and acid
20 NaBH(OAc)₃. This reaction can be used to prepare the compounds encompassed in general structure 2-E.

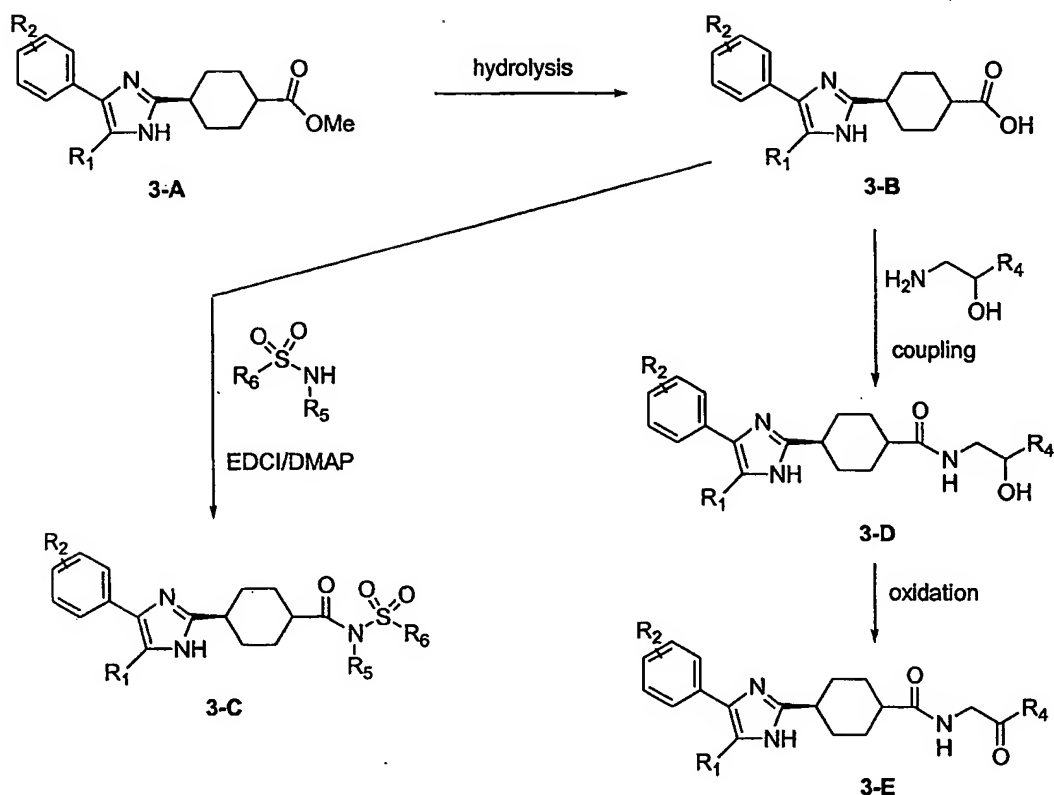
In Scheme 3, "hydrolysis" refers to the synthetic transformation wherein an ester moiety is converted to a carboxylic acid group. Such a transformation can be used to prepare compounds of general structure 3-B. The reagents suitable for carrying
25 out this transformation are well known to those skilled in the art of organic synthesis and include, but are not limited to, KOH, NaOH and HCl.

In Scheme 3, "oxidation" refers to the synthetic transformation wherein an alcohol moiety is converted to a ketone group. Such a transformation can be used to prepare compounds of general structure 3-E. Oxidation methods are well known to
30 those skilled in the art of organic synthesis and include, but are not limited to, Swern oxidation and Dess-Martin oxidation.

- 29 -

Scheme 1Scheme 2

- 30 -
Scheme 3



5 In certain situations, the compounds of this invention may contain one or more asymmetric carbon atoms, so that the compounds can exist in different stereoisomeric forms. These compounds can be, for example, racemates or optically active forms. As noted above, all stereoisomers are encompassed by the present invention. Nonetheless, it may be desirable to obtain single enantiomers (*i.e.*, optically

10 active forms). Standard methods for preparing single enantiomers include asymmetric synthesis and resolution of the racemates. Resolution of the racemates can be accomplished, for example, by conventional methods such as crystallization in the presence of a resolving agent, or chromatography using, for example a chiral HPLC column.

15 As noted above, the present invention encompasses pharmaceutically acceptable salts of the compounds described herein. As used herein, a "pharmaceutically acceptable salt" is an acid or base salt that is generally considered in the art to be suitable for use in contact with the tissues of human beings or animals without excessive toxicity, irritation, allergic response, or other problem or

- 31 -

complication. Such salts include mineral and organic acid salts of basic residues such as amines, as well as alkali or organic salts of acidic residues such as carboxylic acids. Specific pharmaceutical salts include, but are not limited to, salts of acids such as hydrochloric, phosphoric, hydrobromic, malic, glycolic, fumaric, sulfuric, sulfamic, sulfanilic, formic, toluenesulfonic, methanesulfonic, ethane disulfonic, 2-hydroxyethylsulfonic, nitric, benzoic, 2-acetoxybenzoic, citric, tartaric, lactic, stearic, salicylic, glutamic, ascorbic, pamoic, succinic, fumaric, maleic, propionic, hydroxymaleic, hydroiodic, phenylacetic, alkanoic such as acetic, $\text{HOOC}-(\text{CH}_2)_n-\text{COOH}$ where n is 0-4, and the like. Similarly, pharmaceutically acceptable cations include, but are not limited to sodium, potassium, calcium, aluminum, lithium and ammonium. Those of ordinary skill in the art will recognize further pharmaceutically acceptable salts for the compounds provided herein, including those listed by *Remington's Pharmaceutical Sciences*, 17th ed., Mack Publishing Company, Easton, PA, p. 1418 (1985). Accordingly, the present disclosure should be construed to include all pharmaceutically acceptable salts of the compounds specifically recited.

A wide variety of synthetic procedures are available for the preparation of pharmaceutically acceptable salts. In general, a pharmaceutically acceptable salt can be synthesized from a parent compound that contains a basic or acidic moiety by any conventional chemical method. Briefly, such salts can be prepared by reacting the free acid or base forms of these compounds with a stoichiometric amount of the appropriate base or acid in water or in an organic solvent, or in a mixture of the two; generally, nonaqueous media like ether, ethyl acetate, ethanol, isopropanol, or acetonitrile are preferred.

The present invention also encompasses prodrugs of the compounds of Formula I, which may be modified (either in routine manipulation or *in vivo*) to generate an active agent encompassed by Formula I. Such prodrugs may be prepared by modifying functional groups present in the compounds in such a way that the modifications are cleaved to the parent compounds. Prodrugs include compounds wherein hydroxy, amine or sulfhydryl groups are bonded to any group that, when administered to a mammalian subject, cleaves to form a free hydroxyl, amino, or sulfhydryl group, respectively. Examples of prodrugs include, but are not limited to, acetate, formate and benzoate derivatives of alcohol and amine functional groups within the compounds provided herein. Preferred prodrugs include acylated derivatives. Those

- 32 -

of ordinary skill in the art will recognize various synthetic methods that may be employed to prepare prodrugs of the compounds provided herein.

Additional moieties may be associated with a compound using any suitable procedure. Covalent linkage may generally be achieved using suitable functional groups (*e.g.*, hydroxyl, carboxyl, sulfhydryl or amino groups) on the compound and the moiety to be attached. For example, a nucleophilic group, such as an amino or sulfhydryl group, on one may be capable of reacting with a carbonyl-containing group, such as an anhydride or an acid halide, or with an alkyl group containing a good leaving group (*e.g.*, a halide) on the other. The use of bifunctional, multifunctional and/or cleavable linkers may also be desirable for certain applications. Such linkers are well known in the art. Compounds associated with carriers may be covalently linked or, preferably, such association does not involve covalent interaction and is achieved by mixing.

Compounds may be radiolabeled by carrying out their synthesis using precursors comprising at least one atom that is a radioisotope (*i.e.*, an isotopically labeled reagent is substituted for a non-isotopically labeled reagent). Numerous radioisotopes are readily available, including isotopes of hydrogen, carbon, nitrogen, oxygen, phosphorous, sulfur, iodine, fluorine and chlorine, such as ^{14}C , ^3H , ^{35}S or ^{125}I . Synthesis of radiolabeled compounds may be conveniently performed by a radioisotope supplier specializing in custom synthesis of radiolabeled probe compounds, such as Amersham Corporation, Arlington Heights, IL; Cambridge Isotope Laboratories, Inc. Andover, MA; SRI International, Menlo Park, CA; Wizard Laboratories, West Sacramento, CA; ChemSyn Laboratories, Lexena, KS; American Radiolabeled Chemicals, Inc., St. Louis, MO; and Moravsek Biochemicals Inc., Brea, CA. Tritium labeled compounds are also conveniently prepared catalytically via platinum-catalyzed exchange in tritiated acetic acid, acid-catalyzed exchange in tritiated trifluoroacetic acid, or heterogeneous-catalyzed exchange with tritium gas. Such preparations may also be performed as a custom radiolabeling by any of the suppliers listed above using the compound as substrate. In addition, certain precursors may be subjected to tritium-halogen exchange with tritium gas, tritium gas reduction of unsaturated bonds, or reduction using sodium borotritide, as appropriate.

- 33 -

Pharmaceutical Compositions

The present invention also provides pharmaceutical compositions comprising NPY5 receptor modulators, together with at least one physiologically acceptable carrier or excipient. Such compositions may comprise, for example, water, buffers (*e.g.*, neutral buffered saline or phosphate buffered saline), ethanol, mineral oil, vegetable oil, dimethylsulfoxide, carbohydrates (*e.g.*, glucose, mannose, sucrose or dextrans), mannitol, proteins, adjuvants, polypeptides or amino acids such as glycine, antioxidants, chelating agents such as EDTA or glutathione and/or preservatives. Preferred pharmaceutical compositions are formulated for oral delivery to humans or other animals (*e.g.*, companion animals such as dogs).

If desired, other active ingredients may also be included. For example, compositions intended for the treatment of eating disorders, particularly obesity and bulimia nervosa, may further comprise an agent such as sibutramine, dextenfluramine, leptin, a growth hormone secretagogue, a melanocortin agonist, a beta-3 agonist, a 5HT-2 agonist, an orexin antagonist, a melanin concentrating hormone antagonist, a galanin antagonist, a CCK agonist, a GLP-1 agonist and/or a corticotropin-releasing hormone agonist or a NPY₁ antagonist.

Pharmaceutical compositions may be formulated for any appropriate manner of administration, including for example, topical, oral, nasal, rectal or parenteral administration. The term parenteral as used herein includes subcutaneous injections, intradermal, intravascular (*e.g.*, intravenous), intramuscular, spinal, intracranial, intrathecal, intraperitoneal injection or like injection or infusion techniques. In certain embodiments, compositions in a form suitable for oral use are preferred. Such forms include, for example, tablets, troches, lozenges, aqueous or oily suspensions, dispersible powders or granules, emulsion, hard or soft capsules, or syrups or elixirs. Within yet other embodiments, compositions of the present invention may be formulated as a lyophilizate.

Compositions intended for oral use may further contain one or more components such as sweetening agents, flavoring agents, coloring agents and preserving agents in order to provide appealing and palatable preparations. Tablets contain the active ingredient(s) in admixture with physiologically acceptable excipients that are suitable for the manufacture of tablets. Such excipients include, for example, inert diluents (*e.g.*, calcium carbonate, sodium carbonate, lactose, calcium phosphate or

- 34 -

sodium phosphate), granulating and disintegrating agents (*e.g.*, corn starch or alginic acid), binding agents (*e.g.*, starch, gelatin or acacia) and lubricating agents (*e.g.*, magnesium stearate, stearic acid or talc). The tablets may be uncoated or they may be coated by known techniques to delay disintegration and absorption in the
5 gastrointestinal tract and thereby provide a sustained action over a longer period. For example, a time delay material such as glyceryl monostearate or glyceryl distearate may be employed.

Formulations for oral use may also be presented as hard gelatin capsules in which the active ingredient is mixed with an inert solid diluent (*e.g.*, calcium
10 carbonate, calcium phosphate or kaolin), or as soft gelatin capsules wherein the active ingredient is mixed with water or an oil medium (*e.g.*, peanut oil, liquid paraffin or olive oil).

Aqueous suspensions contain the active materials in admixture with excipients suitable for the manufacture of aqueous suspensions. Such excipients are
15 suspending agents (*e.g.*, sodium carboxymethylcellulose, methylcellulose, hydropropylmethylcellulose, sodium alginate, polyvinylpyrrolidone, gum tragacanth and gum acacia); and dispersing or wetting agents (*e.g.*, naturally-occurring phosphatides such as lecithin, condensation products of an alkylene oxide with fatty acids such as polyoxyethylene stearate, condensation products of ethylene oxide with long chain
20 aliphatic alcohols such as heptadecaethyleneoxycetanol, condensation products of ethylene oxide with partial esters derived from fatty acids and a hexitol such as polyoxyethylene sorbitol monooleate, or condensation products of ethylene oxide with partial esters derived from fatty acids and hexitol anhydrides such as polyethylene sorbitan monooleate). Aqueous suspensions may also contain one or more
25 preservatives, for example ethyl, or n-propyl p-hydroxybenzoate, one or more coloring agents, one or more flavoring agents, and/or one or more sweetening agents, such as sucrose or saccharin.

Oily suspensions may be formulated by suspending the active ingredients in a vegetable oil (*e.g.*, arachis oil, olive oil, sesame oil or coconut oil), or in a mineral
30 oil such as liquid paraffin. The oily suspensions may also contain a thickening agent such as beeswax, hard paraffin or cetyl alcohol. Sweetening agents, such as those set forth above, and/or flavoring agents may be added to provide palatable oral

- 35 -

preparations. Such suspensions may be preserved by the addition of an anti-oxidant such as ascorbic acid.

Dispersible powders and granules suitable for preparation of an aqueous suspension by the addition of water provide the active ingredient in admixture with a dispersing or wetting agent, suspending agent and/or one or more preservatives. Suitable dispersing or wetting agents and suspending agents are exemplified by those already mentioned above. Additional excipients, such as sweetening, flavoring and/or coloring agents, may also be present.

Pharmaceutical compositions may also be in the form of oil-in-water emulsions. The oily phase may be a vegetable oil (*e.g.*, olive oil or arachis oil), or a mineral oil (*e.g.*, liquid paraffin) or mixtures thereof. Suitable emulsifying agents may be naturally-occurring gums (*e.g.*, gum acacia or gum tragacanth), naturally-occurring phosphatides (*e.g.*, soy bean, lecithin and esters or partial esters derived from fatty acids and hexitol), anhydrides (*e.g.*, sorbitan monoleate) and condensation products of partial esters derived from fatty acids and hexitol with ethylene oxide (*e.g.*, polyoxyethylene sorbitan monoleate). The emulsions may also contain sweetening and/or flavoring agents.

Syrups and elixirs may be formulated with sweetening agents, such as glycerol, propylene glycol, sorbitol or sucrose. Such formulations may also comprise one or more demulcents, preservatives, flavoring agents and/or coloring agents.

A pharmaceutical composition may be prepared as a sterile injectable aqueous or oleaginous suspension. The modulator, depending on the vehicle and concentration used, can either be suspended or dissolved in the vehicle. Such a composition may be formulated according to the known art using suitable dispersing, wetting agents and/or suspending agents, such as those mentioned above. Among the acceptable vehicles and solvents that may be employed are water, 1,3-butanediol, Ringer's solution and isotonic sodium chloride solution. In addition, sterile, fixed oils may be used as a solvent or suspending medium. For this purpose any bland fixed oil may be employed, including synthetic mono- or diglycerides. In addition, fatty acids such as oleic acid find use in the preparation of injectable compositions, and adjuvants such as local anesthetics, preservatives and/or buffering agents can also be dissolved in the vehicle.

- 36 -

Modulators may also be prepared in the form of suppositories (*e.g.*, for rectal administration). Such compositions can be prepared by mixing the drug with a suitable non-irritating excipient that is solid at ordinary temperatures but liquid at the rectal temperature and will therefore melt in the rectum to release the drug. Suitable excipients include, for example, cocoa butter and polyethylene glycols.

For administration to non-human animals, the composition may also be added to animal feed or drinking water. It may be convenient to formulate animal feed and drinking water compositions so that the animal takes in an appropriate quantity of the composition along with its diet. It may also be convenient to present the composition as a premix for addition to feed or drinking water.

Pharmaceutical compositions may be formulated as sustained release formulations (*i.e.*, a formulation such as a capsule that effects a slow release of modulator following administration). Such formulations may generally be prepared using well known technology and administered by, for example, oral, rectal or subcutaneous implantation, or by implantation at the desired target site. Carriers for use within such formulations are biocompatible, and may also be biodegradable; preferably the formulation provides a relatively constant level of modulator release. The amount of modulator contained within a sustained release formulation depends upon the site of implantation, the rate and expected duration of release and the nature of the condition to be treated or prevented.

Modulators are generally present within a pharmaceutical composition in a therapeutically effective amount. A therapeutically effective amount is an amount that results in a discernible patient benefit, such as increased healing of a disease or disorder associated with pathogenic NPY5 receptor activation, as described herein. A preferred concentration is one sufficient to inhibit the binding of ligand (*i.e.*, NPY and/or PYY) to NPY5 receptor *in vitro*. Compositions providing dosage levels ranging from about 0.1 mg to about 140 mg per kilogram of body weight per day are preferred (about 0.5 mg to about 7 g per human patient per day). The amount of active ingredient that may be combined with the carrier materials to produce a single dosage form will vary depending upon the host treated and the particular mode of administration. Dosage unit forms will generally contain between from about 1 mg to about 500 mg of an active ingredient. It will be understood, however, that the optimal dose for any particular patient will depend upon a variety of factors, including the activity of the specific

- 37 -

compound employed; the age, body weight, general health, sex and diet of the patient; the time and route of administration; the rate of excretion; any simultaneous treatment, such as a drug combination; and the type and severity of the particular disease undergoing treatment. Optimal dosages may be established using routine testing, and
5 procedures that are well known in the art.

Pharmaceutical compositions may be packaged for treating disorders responsive to NPY5 receptor modulation (*e.g.*, treatment of eating disorders such as obesity or bulimia, psychiatric disorders, cardiovascular disorders such as hypertension or diabetes). Packaged pharmaceutical compositions generally include a container
10 holding a therapeutically effective amount of at least one NPY5 receptor modulator as described herein and instructions (*e.g.*, labeling) indicating that the contained composition is to be used for treating a disorder associated with NPY5 receptor activation in the patient.

Methods of Use

15 Within certain aspects, the present invention provides methods for inhibiting the development of a disease or disorder associated with NPY5 receptor activation. In other words, therapeutic methods provided herein may be used to treat an existing disease or disorder, or may be used to prevent or delay the onset of such a disease in a patient who is free of a detectable disease or disorder that is associated with
20 NPY5 receptor activation. As used herein, a disease or disorder is "associated with NPY5 receptor activation" if it is characterized by inappropriate stimulation of NPY5 receptor, regardless of the actual amount of NPY present locally. Such conditions include, for example, eating disorders (such as obesity, anorexia, bulimia and metabolic disorders), diseases related to the central nervous system (such as psychiatric disorders),
25 diseases related to abnormal hormone release (such as diabetes) and cardiovascular disorders. Diseases related to the central nervous system include cerebral infarction, neurodegeneration, epilepsy, stroke and conditions related to stroke, cerebral vasospasm and hemorrhage, depression, anxiety, schizophrenia and dementia, as well as conditions related to pain or nociception. Diseases related to abnormal hormone release include
30 conditions associated with abnormal release of leutinizing hormone, growth hormone, insulin and prolactin. Cardiovascular disorders include any disorders or diseases pertaining to the heart, blood vessels or the renal system, such as hypertension, vasospasm, heart failure, shock, cardiac hypertrophy increased blood pressure, angina,

- 38 -

myocardial infarction, sudden cardiac death, arrhythmia, peripheral vascular disease, and abnormal renal conditions such as impaired flow of fluid, abnormal mass transport or renal failure. Other diseases and disorders associated with NPY5 receptor activation include conditions related to increased sympathetic nerve activity (e.g., during or after coronary artery surgery, and operations and surgery in the gastrointestinal tract); diseases related to abnormal gastrointestinal motility and secretion (such as different forms of ileus, urinary incontinence and Crohn's disease); diseases related to sexual dysfunction and reproductive disorders; conditions or disorders associated with inflammation; and respiratory diseases, such as asthma and conditions related to asthma and bronchoconstriction. The above conditions may be diagnosed and monitored using criteria that have been established in the art. Patients may include humans, domesticated companion animals (pets, such as dogs) and livestock animals, with dosages and treatment regimes as described above.

Frequency of dosage may vary depending on the compound used and the particular disease to be treated or prevented. In general, for treatment of most disorders, a dosage regimen of 4 times daily or less is preferred. For the treatment of eating disorders, including obesity, a dosage regimen of 1 or 2 times daily is particularly preferred. For the treatment of impotence a single dose that rapidly reaches effective concentrations is desirable. It will be understood, however, that the specific dose level for any particular patient will depend upon a variety of factors including the activity of the specific compound employed, the age, body weight, general health, sex, diet, time of administration, route of administration, and rate of excretion, drug combination and the severity of the particular disease undergoing therapy. In general, the use of the minimum dosage that is sufficient to provide effective therapy is preferred. Patients may generally be monitored for therapeutic effectiveness using assays suitable for the condition being treated or prevented, which will be familiar to those of ordinary skill in the art.

Within separate aspects, the present invention provides a variety of *in vitro* uses for the compounds provided herein. For example, such compounds may be used as probes for the detection and localization of NPY5 receptors, in samples such as tissue sections, as positive controls in assays for receptor activity, as standards and reagents for determining the ability of a candidate agent to bind to NPY5 receptor, or as radiotracers for positron emission tomography (PET) imaging or for single photon

- 39 -

emission computerized tomography (SPECT). Such assays can be used to characterize NPY5 receptors in living subjects.

Within methods for determining the presence or absence of NPY5 receptor in a sample, a sample may be incubated with a compound as provided herein under conditions that permit binding of the compound to NPY5 receptor. The amount of compound bound to NPY5 receptor in the sample is then detected. For example, a compound may be labeled using any of a variety of well known techniques (e.g., radiolabeled with a radionuclide such as tritium, as described herein), and incubated with the sample (which may be, for example, a preparation of cultured cells, a tissue preparation or a fraction thereof). A suitable incubation time may generally be determined by assaying the level of binding that occurs over a period of time. Following incubation, unbound compound is removed, and bound compound detected using any method for the label employed (e.g., autoradiography or scintillation counting for radiolabeled compounds; spectroscopic methods may be used to detect dyes, luminescent groups and fluorescent groups). Detection assays, including receptor autoradiography (receptor mapping) of NPY5 receptors in cultured cells or tissue samples may be performed as described by Kuhar in sections 8.1.1 to 8.1.9 of Current Protocols in Pharmacology (1998) John Wiley & Sons, New York.

Modulators provided herein may also be used within a variety of well known cell culture and cell separation methods. For example, modulators may be linked to the interior surface of a tissue culture plate or other cell culture support, for use in immobilizing NPY5-expressing cells for screens, assays and growth in culture. Such linkage may be performed by any suitable technique, such as the methods described above, as well as other standard techniques. Modulators may also be used to facilitate cell identification and sorting *in vitro*, permitting the selection of cells expressing NPY5. Preferably, the modulator(s) for use in such methods are labeled as described herein. Within one preferred embodiment, a modulator linked to a fluorescent marker, such as fluorescein, is contacted with the cells, which are then analyzed by fluorescence activated cell sorting (FACS).

Within other aspects, methods are provided for modulating binding of an NPY5 receptor ligand (such as NPY or PYY) to NPY5 receptor *in vitro* or *in vivo*, comprising contacting a sufficient amount of NPY5 receptor with a modulator provided herein, under conditions suitable for binding of NPY5 to the receptor. Preferably,

- 40 -

within such methods, NPY and/or PYY binding to receptor is inhibited by the modulator. The NPY₅ receptor may be present in solution, in a cultured or isolated cell preparation or within a patient. In general, the amount of compound contacted with the receptor *in vivo* should be sufficient to modulate NPY binding to NPY₅ receptor *in vitro* within, for example, a ligand binding assay as described in Example 11. NPY₅ receptors used to determine *in vitro* binding may be obtained from a variety of sources, for example from preparations of rat brain or from cells expressing cloned human NPY₅ receptors.

Also provided herein are methods for modulating the signal-transducing activity of NPY₅ receptor, by contacting an NPY₅ receptor, either *in vivo* or *in vitro*, with a sufficient amount of an NPY₅ receptor modulator as described above, under conditions suitable for binding of NPY to NPY₅ receptor. The NPY₅ receptor may be present in solution, in a cultured or isolated cell preparation or within a patient. In general, the amount of a modulator that is sufficient to alter the signal-transducing activity of NPY₅ receptor may be determined via a NPY₅ receptor signal transduction assay, such as the assay described in Example 12.

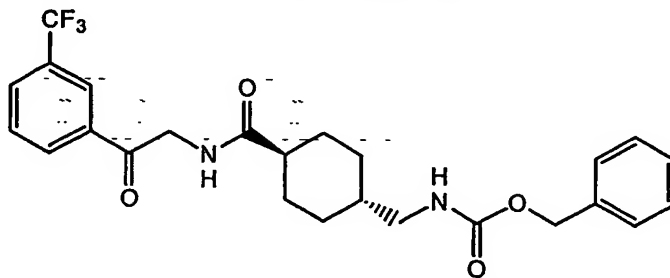
The following Examples are offered by way of illustration and not by way of limitation. Unless otherwise specified all reagents and solvent are of standard commercial grade and are used without further purification.

Examples

Example 1

Preparation of Cyclopentyl-[4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl]-amine

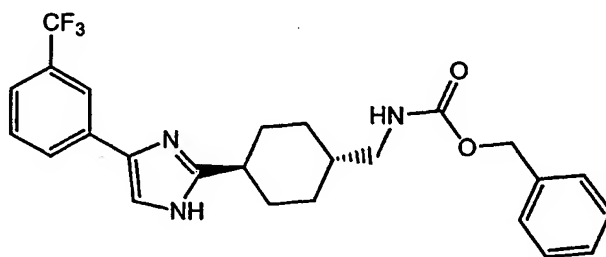
This Example illustrates the preparation of a representative 2-cyclohexyl-4-phenyl-1H-imidazole derivative having the structure:



A. To a solution of 2-Amino-1-(3-trifluoromethyl-phenyl)-ethanone hydrochloride (3.4 g, 0.0144 mol (prepared according to the procedure of Leclerc and

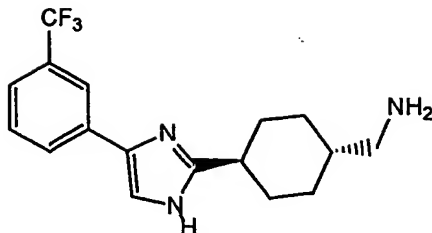
- 41 -

- Bizec (1980) *J. Med. Chem.* 23:783-744) and 4-(Benzyloxycarbonylamino-methyl)-cyclohexanecarboxylic acid (4.2 g, 0.0144 mol; for a preparation of this reagent see e.g., Svahn et. al. (1986) *J. Med. Chem.* 29:448-453) in DMF, Benzotriazol-1-yloxytris(dimethylamino)phosphonium hexafluorophosphate (BOP reagent, 6.4 g, 0.0144 mol) was added, followed by triethylamine (4.0 mL, 0.0289 mol). The reaction was stirred overnight and then poured into water to produce a thick precipitate. The solid was collected on a sintered glass funnel and washed consecutively with H₂O, 3N HCl (2x), H₂O, NaHCO₃ solution (2x) and H₂O. The filter cake was air dried, added to a 500 mL round bottom flask with toluene and concentrated under reduced pressure.
- Additional toluene (200 mL) was added and the mixture was concentrated again under reduced pressure to yield {4-[2-Oxo-2-(3-trifluoromethyl-phenyl)-ethylcarbamoyl]-cyclohexylmethyl}-carbamic acid benzyl ester as a solid. ¹H NMR (DMSO-d₆): δ 0.8-0.93 (m, 2H), 1.2-1.37 (m, 3H), 1.71 (br s, 4H), 2.14 (t, 1H), 2.84 (br s, 2H), 4.57 (br s, 2H), 4.99 (s, 2H), 7.2-7.38 (m, 5H), 7.77(t, 1H), 8.01 (d, J=8.0 Hz, 1H), 8.14-8.26 (m, 3H). MS 477 (M+H)⁺

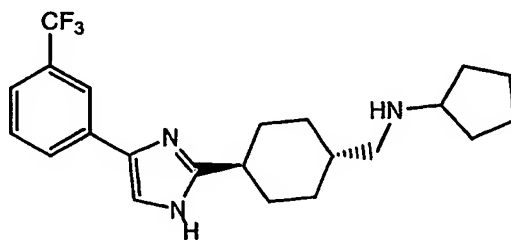


- B. Ammonium acetate (26 g, 0.34 mol) was added to a solution of {4-[2-Oxo-2-(3-trifluoromethyl-phenyl)-ethylcarbamoyl]-cyclohexylmethyl}-carbamic acid benzyl ester (5.4 g, 0.0113 mol) in acetic acid. The resulting homogeneous mixture was refluxed overnight and then concentrated under reduced pressure. The residue was partitioned between 10% NaOH and EtOAc (1:1) and the EtOAc layer was washed with 10% NaOH (3x) and brine. The EtOAc was dried (Na₂SO₄) and concentrated under reduced pressure to give {4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-carbamic acid benzyl ester. ¹H NMR (CDCl₃): δ 1.02-1.18 (m, 2H), 1.46-1.62 (m, 3H), 1.86-1.96 (m, 2H), 2.12-2.21 (m, 2H), 2.74 (m, 1H), 3.10 (t, J=6 Hz, 2H), 4.91 (t, 1H, N-H), 5.10 (s, 2H), 7.22 (s, 1H), 7.32-7.37 (m, 5H), 7.45 (d, J=4.9 Hz, 2H), 7.87 (m, 1H), 7.95 (s, 1H). MS 458 (M+H)⁺

- 42 -



C. An ethanolic solution of {4-[4-(3-Trifluoromethyl-phenyl)-1*H*-imidazol-2-yl]-cyclohexylmethyl}-carbamic acid benzyl ester (4.15 g, 0.0091 mol) and 10% Pd/Carbon (400 mg) was shaken overnight under 55 psi of hydrogen in a Paar apparatus. The mixture was filtered through celite and concentrated under reduced pressure to give a foam, which was triturated with EtOAc/Hexanes/MeOH to produce C-4-[4-(3-trifluoromethyl-phenyl)-1*H*-imidazole-2-yl]-cyclohexyl}-methylamine as a filterable solid. ¹H NMR (DMSO-*d*₆): δ 0.93-1.05 (m, 2H), 1.30-1.4 (m, 2H), 1.42-1.55 (m, 2H), 1.85 (d, *J* = 12.8 Hz, 2H), 1.99 (d, *J* = 11.6 Hz, 2H), 2.4-2.5 (m, 1H), 2.56-2.66 (m, 1H), 7.43-7.48 (m, 1H), 7.50-7.55 (m, 1H), 7.63 (s, 1H), 7.99 (d, *J* = 7.9 Hz, 1H), 8.02 (s, 1H). MS 324 (M+H)⁺

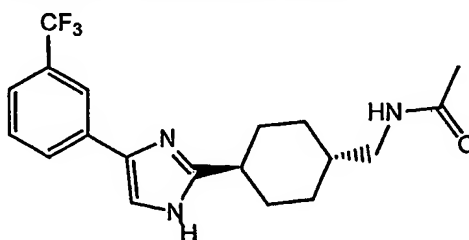


D. C-4-[4-(3-trifluoromethyl-phenyl)-1*H*-imidazole-2-yl]-cyclohexyl}-methylamine (217 mg, 0.672 mmol) was dissolved in a mixture of dichloroethane and THF (~2:1). Cyclopentanone (62 mg, 0.74 mmol) was added to the reaction mixture, followed by acetic acid (0.672 mmol). Sodium triacetoxyborohydride was added and the reaction was stirred overnight at room temperature. The reaction mixture was concentrated under reduced pressure and partitioned between ethyl acetate and 10% NaOH. The ethyl acetate layer was dried (Na₂SO₄), concentrated under reduced pressure, and purified using preparative plate chromatography (MeOH/CH₂Cl₂ eluent) to generate cyclopentyl-{4-[4-(3-trifluoromethyl-phenyl)-1*H*-imidazol-2-yl]-cyclohexylmethyl}-amine.

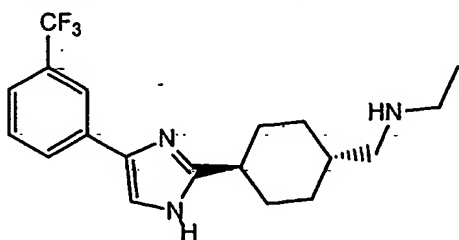
- 43 -

Example 2Preparation of Ethyl-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazole-2-yl]cyclohexylmethyl}-amine

This Example illustrates the preparation of a representative 2-cyclohexyl-4-phenyl-1H-imidazole derivative having the structure:



- A. C-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazole-2-yl]-cyclohexyl}-methylamine (326 mg, 1.0 mmol, from Example 1C) and TEA (102 mg, 1.0 mmol) were dissolved in CH₂Cl₂/THF (1:1) and brought to 0°C with an ice bath. Acetyl chloride (79 mg, 1.0 mmol) was added to the reaction mixture slowly and the ice bath was removed. After 2 hours, the solution was diluted with CH₂Cl₂, added to a separatory funnel and washed with 10% NaOH (3X). The solution was dried (Na₂SO₄), concentrated under reduced pressure, and the residue purified using flash chromatography (5%→10% MeOH/CH₂Cl₂) to give (N-{4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-acetamide.



- B. To a solution of (N-{4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-acetamide (207 mg, 0.57 mmol, from Example 2A) in THF, 3 equivalents of borane dimethylsulfide (2M in THF) was added, and the solution stirred overnight at room temperature. One volume of MeOH was added to the reaction mixture, and the mixture was concentrated under reduced pressure. 50 mL of 3N HCl was added, and the mixture was warmed for 5 minutes. After washing the solution with ether, the solution was made basic with NaOH and extracted with ethyl acetate. The solution was dried (Na₂SO₄), concentrated under reduced pressure, and

- 44 -

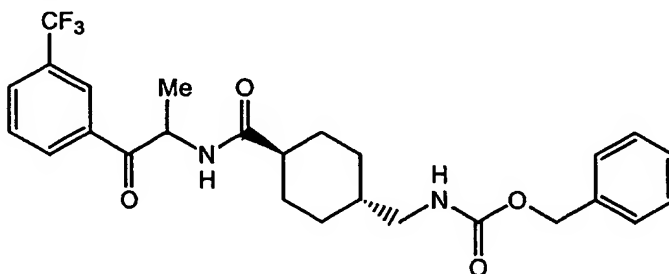
purified using preparative plate chromatography to give Ethyl-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazole-2-yl]cyclohexylmethyl}-amine.

Example 3

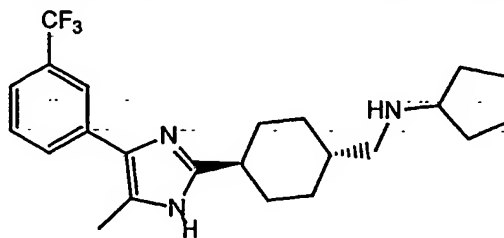
Preparation of Cyclopentyl-{4-[5-methyl-4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-amine

5

This Example illustrates the preparation of a representative 2-cyclohexyl-4-phenyl-1H-imidazole derivative having the structure:



- A. This compound was prepared using a procedure similar to the one described by Moriya et. al. (J. Med. Chem. (1986) 29:333-341) for the alkylation of α -(N-acylamino) ketones: {4-[2-Oxo-2-(3-trifluoromethyl-phenyl)-ethylcarbamoyl]-cyclohexylmethyl}-carbamic acid benzyl ester (From Example 1A; 584 mg, 1.23 mmol) was dissolved in dry DMF and brought to -40°C using a dry ice/acetonitrile cooling bath. NaH (61 mg, 1.53 mmol, 60% dispersion in oil) was added in one portion and the mixture was stirred for 15 minutes. MeI (192 mg, 1.35 mmol) was added all at once, the cooling bath was removed and the reaction mixture was allowed to come to room temperature. An excess of saturated aqueous NH_4Cl solution was added and the resulting mixture extracted with EtOAc. The EtOAc layer was washed sequentially with 3N HCl, NaHCO_3 (sat'd), and brine. The solution was dried (Na_2SO_4) and concentrated under reduced pressure to give {4-[1-Methyl-2-oxo-2-(3-trifluoromethyl-phenyl)-ethylcarbamoyl]-cyclohexylmethyl}-carbamic acid benzyl ester.



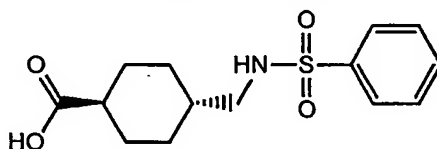
- 45 -

B. Using procedures analogous to those described in Examples 1B to 1D, the compound from Example 3A was converted to Cyclopentyl-{4-[5-methyl-4-(3-trifluoromethyl-phenyl)-1*H*-imidazol-2-yl]-cyclohexylmethyl}-amine.

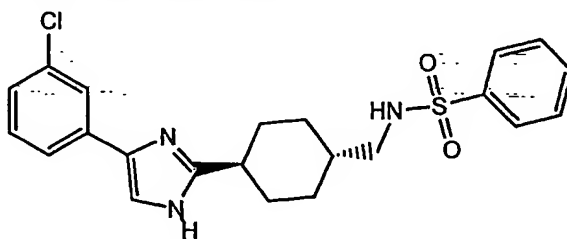
Example 4

5 Preparation of *N*-{4-[4-(3-Chloro-phenyl)-1*H*-imidazol-2-yl]cyclohexylmethyl}-benzenesulfonamide

This Example illustrates the preparation of a representative 2-cyclohexyl-4-phenyl-1*H*-imidazole derivative having the structure:...



- 10 A. *trans*-4-(Aminomethyl)cyclohexane carboxylic acid (15.2 g, 0.097 mol) was dissolved in a 2N NaOH solution (96.5 mL, 2 equivalents). The solution was brought to 10°C, benzenesulfonyl chloride (17.1 g, 0.097 mol) was added dropwise and then the mixture was stirred overnight at room temperature. The mixture was diluted with water, the solution was washed with ether, and then the aqueous
- 15 portion was acidified with 12N HCl to give an oily precipitate. The precipitate was extracted with CH₂Cl₂, and the extracts were dried (Na₂SO₄) and concentrated under reduced pressure to give *trans*-4-(Benzenesulfonylamino-methyl)-cyclohexanecarboxylic acid as a white solid.

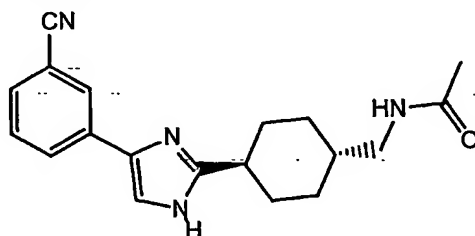


- 20 B. Using procedures analogous to those described in Examples 1A and 1B, the compound from Example 5A was converted to *N*-{4-[4-(3-Chloro-phenyl)-1*H*-imidazol-2-yl]cyclohexylmethyl}-benzenesulfonamide.

- 46 -

Example 5Preparation of *N*-{4-[4-(3-Cyano-phenyl)-1*H*-imidazol-2-yl]-cyclohexylmethyl}-acetamide

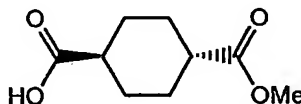
This Example illustrates the preparation of a representative 2-cyclohexyl-4-phenyl-1*H*-imidazole derivative having the structure:



A. A solution of *N*-{4-[4-(3-Bromo-phenyl)-1*H*-imidazol-2-yl]-cyclohexylmethyl}-acetamide (54 mg, 0.14 mmol, prepared using procedures described above), ZnCN_2 (17 mg, 0.14 mmol), and tetrakis(triphenylphosphine)palladium(0) (15 mg) was heated to 150°C for 8 hours. The solution was cooled, diluted with water and extracted with ethyl acetate. The ethyl acetate was washed with 10% NaOH (3X) and brine, dried (Na_2SO_4) and concentrated under reduced pressure. The crude product was purified using preparative plate chromatography (MeOH/ CH_2Cl_2) to give *N*-{4-[4-(3-Cyano-phenyl)-1*H*-imidazol-2-yl]-cyclohexylmethyl}-acetamide.

Example 6Preparation of *trans*-*N*-[(4-methylphenyl)sulfonyl](4-{4-[3-(trifluoromethyl)phenyl]imidazol-2-yl}cyclohexyl)carboxamide

This Example illustrates the preparation of a representative 2-cyclohexyl-4-phenyl-1*H*-imidazole derivative having the structure:

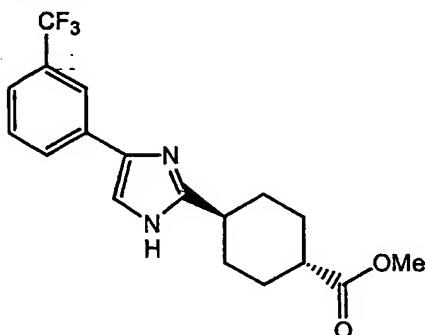


20

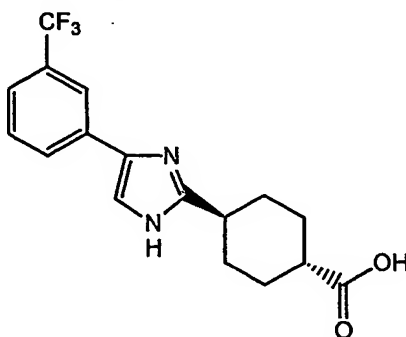
A. *trans*-Cyclohexane-1,4-dicarboxylic acid dimethyl ester (34.2 g, 0.171 mol) was dissolved in 400 mL of anhydrous methanol and the solution brought to reflux. Slowly (dropwise), a solution of KOH (11.2 g, 0.171 mol, 85% purity) in dry methanol was added to the refluxing solution. The solution continued to reflux for 5 hours, and was then cooled and concentrated under reduced pressure to give a white solid. Water was added to dissolve most of the solid, and the aqueous mixture was washed with ether (3X) in a separatory funnel. The aqueous layer was brought to pH

- 47 -

~6, the resulting white precipitate was collected via filtration and washed (1X) with a small amount of water. Air drying afforded *trans*-cyclohexane-1,4-dicarboxylic acid monomethyl ester as a white solid.



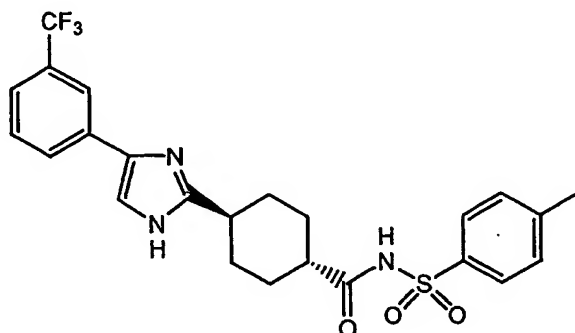
- 5 B. The solution of 2-amino-1-(3-trifluoromethyl-phenyl)-ethanone hydrochloride (50.2 mmol), *trans*-cyclohexane-1,4-dicarboxylic acid monomethyl ester (50.2 mmol), triethylamine (100.4 mmol) and BOP (50.2 mmol) in DMF (100 mL) was stirred overnight at room temperature. The mixture was diluted with water, extracted with EtOAc, washed with saturated NaHCO₃ and brine, dried over Na₂SO₄, and
- 10 concentrated under vacuum. The residue was triturated with ether and the precipitate was collected. The precipitate was mixed with NH₄OAc (930 mmol) and 200 ml of AcOH and refluxed overnight. The excess AcOH was removed and purification by flash chromatography (95:5 CH₂Cl₂/MeOH) gave *trans*-4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexyl-carboxylic acid methyl ester.



- 15
- C. To a solution of *trans*-4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylcarboxylic acid methyl ester (13.4 mmol) in 50 ml of THF was added LiOH (80.3 mmol) in 30 ml of H₂O. The mixture was stirred at 60°C for 2 hours, then concentrated, diluted with H₂O and washed with ether. The aqueous layer
- 20 was acidified to pH 5-6, extracted with EtOAc, dried over Na₂SO₄, and concentrated to

- 48 -

give *trans*-4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexyl-carboxylic acid.

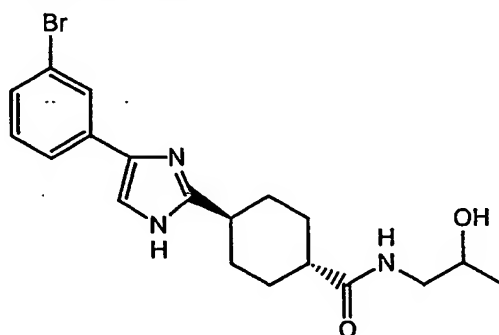


- D. To an ice-water cooled mixture of *trans*-4-{4-[3-(trifluoromethyl)phenyl]imidazol-2-yl}cyclohexanecarboxylic acid (1.48 mmol), 4-methylbenzenesulfonamide (1.48 mmol), and DMAP (1.48 mmol) in dichloromethane (10 mL) under nitrogen, EDC (1.48 mmol) was added. The mixture was stirred at room temperature overnight, concentrated and chromatographed on silica gel (19:1 EtOAc-MeOH eluent) to give *trans*-N-[(4-methylphenyl)sulfonyl](4-{4-[3-(trifluoromethyl)phenyl]-imidazol-2-yl}cyclohexyl) carboxamide.

Example 7

Preparation of *trans*-4-[4-(3-bromo-phenyl)-1H-imidazol-2-yl]-cyclohexane-carboxylic acid (2-hydroxy-propyl) amide

- This Example illustrates the preparation of a representative 2-cyclohexyl-4-phenyl-1H-imidazole derivative having the structure:



- The solution of *trans*-4-[4-(3-bromo-phenyl)-1H-imidazol-2-yl]-cyclohexyl-carboxylic acid (1.22 mmol), 2-hydroxy-propylamine (3.65 mmol) and BOP (1.22 mmol) in DMF (6 mL) was stirred overnight at room temperature. The mixture was diluted with water, extracted with EtOAc, washed with saturated NaHCO₃, dried over Na₂SO₄, and concentrated under vacuum. The residue was purified with flash

- 49 -

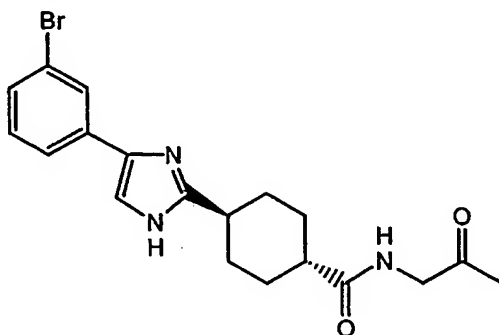
chromatography (90:10:1 CH₂Cl₂/MeOH/NH₄OH) to give *trans*-4-[4-(3-bromo-phenyl)-1H-imidazol-2-yl]-cyclohexane-carboxylic acid (2-hydroxy-propyl) amide.

Example 8

Preparation of *trans*-4-[4-(3-bromo-phenyl)-1H-imidazol-2-yl]-cyclohexane-carboxylic acid (2-oxo-propyl) amide

5

This Example illustrates the preparation of a representative 2-cyclohexyl-4-phenyl-1H-imidazole derivative having the structure:



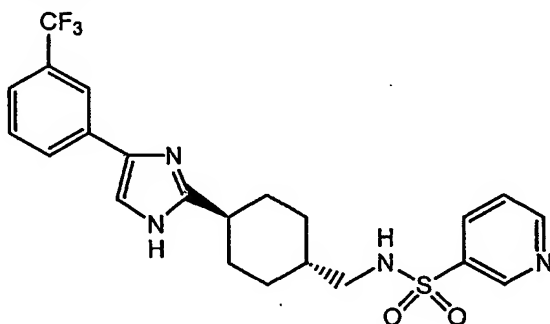
To a cooled solution (-78°C to -60°C) of oxalyl chloride (0.12 mmol) in
10 CH₂Cl₂ (5 mL), DMSO (0.25 mmol) was added dropwise. The mixture was stirred for 2 minutes, and then *trans*-4-[4-(3-bromo-phenyl)-1H-imidazol-2-yl]-cyclohexane-carboxylic acid (2-hydroxy-propyl) amide (0.12 mmol) in 2 ml of CH₂Cl₂ was added. The mixture was stirred for 15 minutes at the same temperature following the addition of TEA (0.60 mmol). The mixture was stirred for another 5 minutes, warmed to room
15 temperature and diluted with water. The organic layer was isolated, dried over Na₂SO₄, concentrated under vacuum, and purified with flash chromatography (90:10:1 CH₂Cl₂/MeOH/NH₄OH) to give *trans*-4-[4-(3-bromo-phenyl)-1H-imidazol-2-yl]-cyclohexane-carboxylic acid (2-oxo-propyl) amide.

- 50 -

Example 9Preparation of Pyridine-3-sulfonic acid *trans*-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-amide

This Example illustrates the preparation of a representative 2-cyclohexyl-

5 4-phenyl-1H-imidazole derivative having the structure:



Pyridine-3-sulfonyl chloride (0.34 mmol) was added to a solution of C-
 {4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexyl}-methanamine (0.31
 mmol) and triethylamine (0.62 mmol) in CH₂Cl₂ (5 mL). The mixture was stirred
 10 overnight at room temperature, diluted with CH₂Cl₂, washed with saturated NaHCO₃,
 dried over Na₂SO₄, and concentrated under vacuum. The residue was purified by flash
 chromatography (90:10 CH₂Cl₂/MeOH) to give Pyridine-3-sulfonic acid *trans*-{4-[4-(3-
 trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-amide...

Example 10Preparation of NPY5 Chimeric Receptors

This Example illustrates the preparation of a representative NPY5
 receptor for use in assays described herein.

A. DNA clones encoding NPY Receptors

Human Y5 receptor was cloned from genomic DNA using a 5' Primer
 20 TTTTGGTTGCTGACAAATGTC (SEQ ID NO:1) and a 3' Primer
 CCTTGGTAAACAGTGAGAATTATTAC (SEQ ID NO:2). The full length PCR
 product was initially cloned into the vector pCR 2.1 (Invitrogen, Carlsbad, CA) and then
 subcloned into pBluescript SK Minus (pBSSKM, Stratagene, La Jolla CA). This was
 designated pNN32.

25 Bases 197 to 1433 of Y1 receptor (Genbank Accession number M88461,
 SEQ ID NO:3) were subcloned into pBSSKM and designated pNN22. A pBSSKM

- 51 -

clone encoding a 5' truncated form of the Y5 receptor was made which deleted the 5' end of the coding region to the Nco I site. This was designated pNN39.

B. Chimeric receptors

(1) hNPY5 Y1IC3 (SEQ ID NO:5)

5 For the IC loop 3 chimera, pNN39 was digested with PstI (located at about residues 748-753 of human Y5 receptor sequence (SEQ ID NO:6) and Bgl II (located at about residues 1130-1135 of the human Y5 receptor sequence) removing bases 753 to 1130.

The portion of IC loop 3 from bases 903-964

10 (TACGCCTAAAAAGGAGAAACAACATGATGGACAAGATGAGAGACAATAAG TACAGGTCCAGT; SEQ ID NO:8) of human Y5 receptor, corresponding to amino acids 236-256 (IRLKRRNNMMDKMRDNKYRSS; SEQ ID NO:9) of the human Y5 receptor amino acid sequence, was inserted into Y5 using the HY1L3S sense oligo (SEQ ID NO:10) and the HY1L3AS antisense oligo (SEQ ID NO:11). A reaction mixture containing the 2 oligos was heated to 100°C and allowed to cool slowly to anneal the oligos. The double stranded annealing product was then ligated into the PstI-Bgl II digested pNN39 to yield plasmid pNN100. The pNN100 insert was then reintroduced into the full-length human Y5 gene (pNN32) at the Cel 2 site and the resulting plasmid was designated pNN42.

20 (2) hNPY5 Y1CT (SEQ ID NO:12)

To add the Y1 C-terminus to Y5, an Eco RI site was added to each gene. For Y1, bases 1173 to 1178 (ACTTCC) of human Y1 receptor (SEQ ID NO:3) were mutated to create an Eco RI site via PCR from forward primer HY1R1 (SEQ ID NO:13) to a T3 reverse primer (priming from the multiple cloning site - "MCS" - of pBSSKM). 25 The Y1 3' tail was then isolated by digesting with Eco RI and Xba (which later enzyme cuts out the Y1 3'tail in the MCS of pBSSKM).

For Y5, bases 1338 to 1343 (GGATTA) of human Y5 receptor were mutated using the PCR reverse primer HY5R1 (SEQ ID NO:14). This primer was paired with a forward primer corresponding to bases 527-551 30 (GCTACTGTCTGGACACTAGGTTTTG; SEQ ID NO:15) of human Y5 receptor, and PCR carried out with the human Y5 coding sequence as template. The resulting PCR band was cut Pst I to the introduced Eco RI site.

- 52 -

pNN39 was then opened Pst I to Xba from the MCS of pBSSKM and the mutated Y5 segment Pst1 to Eco RI was mixed with the mutated Y1 3' fragment Eco RI to Xba from the MCS to set up a three-way ligation. The resulting mutated gene fragment was then introduced into the full-length Y5 gene at the Bgl 2 site.

5 (3) hNPY₅ Y1IC3/ Y1CT (SEQ ID NO:16)

The IC loop 3 + CT tail exchange was obtained by combining the above 2 mutant genes in the following manner. Full length hY5 (pNN32) was digested with Cel 2 (located at about residues 619-625 of human Y5 receptor) and Xba in the vector MCS. The loop 3 mutation pNN42 fragment Cel II to Bgl II was combined with the CT mutation pNN43 fragment Bgl II to Xba from the MCS resulting in pNN44. pNN44 encodes a human chimeric NPY₅/NPY₁ NPY receptor, consisting of N-terminal amino acids 1-442 of the human NPY₅ receptor and C-terminal amino acids 328-384 of the human NPY₁ receptor. The amino acid sequence of this chimeric receptor, referred to herein as hNPY₅ Y1IC3/ Y1CT is shown in SEQ ID NO17:).

15 PNN44 was then digested with Kpn I and Xba I and subcloned into the commercial expression vector pBacPAK9 (Clontech, Palo Alto, CA) for expression in SF9 cells...

C. Baculoviral Preparations

The Baculoviral expression vector was co-transfected along with
20 BACULOGOLD DNA (BD PharMingen, San Diego, CA) into Sf9 cells. The Sf9 cell culture supernatant was harvested three days post-transfection. The recombinant virus-containing supernatant was serially diluted in Hink's TNM-FH insect medium (JRH Biosciences, Kansas City) supplemented Grace's salts and with 4.1mM L-Gln, 3.3 g/L LAH, 3.3 g/L ultrafiltered yeastolate and 10% heat-inactivated fetal bovine serum
25 (hereinafter "insect medium") and plaque assayed for recombinant plaques. After four days, recombinant plaques were selected and harvested into 1 ml of insect medium for amplification. Each 1 ml volume of recombinant baculovirus (at passage 0) was used to infect a separate T25 flask containing 2 x 10⁶ Sf9 cells in 5 mls of insect medium. After five days of incubation at 27°C, supernatant medium was harvested from each of the
30 T25 infections for use as passage 1 inoculum. Two of the seven recombinant baculoviral clones were then chosen for a second round of amplification, using 1 ml of passage 1 stock to infect 1 x 10⁸ cells in 100 ml of insect medium divided into 2 T175 flasks. Forty-eight hours post infection, passage 2 medium from each 100ml prep was

- 53 -

harvested and plaque assayed for titer. The cell pellets from the second round of amplification were assayed by affinity binding as described below to verify recombinant receptor expression. A third round of amplification was then initiated using an M.O.I. of 0.1 to infect a liter of Sf9 cells. Forty hours post-infection the supernatant medium
5 was harvested to yield passage 3 baculoviral stock and the cell pellet assayed for affinity binding. Titer of the passage 3 baculoviral stock was determined by plaque assay and an M.O.I. and Incubation Time Course experiment was carried out to determine conditions for optimal receptor expression. Results from the receptor optimization experiment show that an M.O.I. of 0.1 and a 72 hour incubation were the ideal infection
10 parameters in order to achieve optimum Y5 receptor expression in up to 1 liter Sf9 cell infection cultures.

Log-phase Sf9 cells infected with recombinant baculovirus encoding the human chimeric NPY₅/NPY₁ NPY receptor designated hNPY₅ Y1IC3/ Y1CT, above, were cultured in insect medium at 27°C. 72 hours post-infection, a sample of cell
15 suspension was analyzed for viability by trypan blue dye exclusion, and the remaining Sf9 cells were harvested via centrifugation (3000 rpm/10 minutes/4°C).

Example 11

Assay for NPY₅ Receptor Binding Activity

This Example illustrates the measurement of binding activity for
20 representative compounds.

The baculovirus-infected Sf9 cells expressing recombinant human chimeric NPY₅/NPY₁ receptor, as described in Example 10, were harvested at 42-48 hours at which time batches of 500 mL of cell suspension were pelleted by centrifugation. Each pellet was re-suspended in 30 mL of homogenization buffer (10
25 mM HEPES, 250 mM sucrose, 0.5 µg/mL leupeptin, 2 µg/mL Aprotinin, 200 µM PMSF and 2.5 mM EDTA, pH 7.4) and homogenized using a POLYTRON homogenizer (setting 5 for 30 seconds). The homogenate was centrifuged at 4°C for 10 minutes at 536 x g to pellet the nuclei. The supernatant containing isolated membranes... was decanted into a clean centrifuge tube, centrifuged in the same buffer at 48,000 x g
30 for 30 minutes at 4°C and resuspended in 30 ml homogenization buffer. This centrifugation and resuspension step was repeated twice. The final pellet was re-suspended in ice cold Dulbecco's PBS containing 5 mM EDTA, and stored in frozen

- 54 -

aliquots at -80°C. The protein concentration of the resulting membrane preparation (P2 preparation) was measured using the Bradford protein assay (Bio-Rad Laboratories, Hercules, CA). By this measure, a 1-liter culture of cells typically yields 50-75 mg of total membrane protein.

- 5 Purified P2 membranes were thawed, centrifuged and washed by PBS and re-suspended by Dounce homogenization (tight pestle) in binding buffer (50 mM Tris(HCl), 5 mM KCl, 120 mM NaCl, 2 mM CaCl₂, 1 mM MgCl₂, 0.1% bovine serum albumin (BSA), pH 7.4). For competition analysis, membranes (5-50µg) were added to polypropylene tubes containing 0.050 nM [¹²⁵I]PYY(porcine) and 2µl test compound in
- 10 DMSO (1µM-4µM final concentration). For saturation binding analysis, membranes (5-50 µg) were added to polypropylene tubes containing 0.010-0.500nM [¹²⁵I]PYY (porcine; New England Nuclear Corp., Boston, MA). Nonspecific binding was determined in the presence of 1 µM NPY (human; American Peptide Co., Sunnyvale, CA) and accounted for less than 10% of total binding. Following a 2 hour incubation at
- 15 room temperature, the reaction was terminated by rapid vacuum filtration. Samples were filtered over presoaked (in 1.0% polyethyleneimine for 2 hours prior to use) GF/C WHATMAN filters and rinsed 2 times with 5 mL cold binding buffer without BSA. Remaining bound radioactivity was measured by gamma counting. K_i and Hill coefficient ("nH") were determined by fitting the Hill equation to the measured values.....
- 20 with the aid of SIGMAPLOT software (SPSS Inc., Chicago). The binding affinity for the compounds provided herein, expressed as a K_i value, ranged from about 0.1 nanomolar to about 10 micromolar. The most active of these compounds have a K_i of less than 100 nanomolar.

Example 12

25 Calcium Mobilization Assay for Determining NPY5 Receptor Modulation

This Example illustrates a representative assay for evaluating the effect of compounds on NPY5 receptor signal transduction.

- Bowes Melanoma cells stably transfected with an expression vector encoding the NPY5/NPY1 chimeric receptor described above were plated at a density of
- 30 26,000 cells/well in FALCON black-walled, clear-bottomed 96-well plates (#3904, BECTON-DICKINSON, Franklin Lakes, NJ) and grown to confluency, approximately 24 hours. The culture medium was emptied from the 96 well plate, and Fluo-3 calcium sensitive dye (Molecular Probes, Eugene, OR) was added to each well (dye solution: 1

- 55 -

mg FLUO-3 AM, 440 μ L DMSO and 440 μ l 20% pluronic acid in DMSO, diluted 1:2, 50 μ l diluted solution per well). Plates were covered with aluminum foil and incubated at 37 C for 1-2 hours. After the incubation, the dye solution was emptied from the plates, cells were washed once in 100 μ l KRH buffer (0.05 mM KCl, 0.115 M NaCl, 9.6 mM NaH_2PO_4 , 0.01 mM MgSO_4 , 25 mM HEPES, pH 7.4) to remove excess dye. After washing, 80 μ l KRH buffer with carbachol/Thomae (KRH buffer containing 1 mM carbachol, 10 micromolar BIBP 3226 (Sigma RBI, St. Louis MO)) was added to each well. Assay plates were incubated in the dark, 20 minutes.

To measure the ability of a test compound to antagonize the response of cells expressing NPY5 receptor to NPY, the EC_{50} of NPY was first determined. An additional 20 μ l of KRH/Thomae buffer and 1 μ l DMSO was added to each well of cells, prepared as described above. 100 μ l human NPY in KRH/Thomae buffer was automatically transferred by the FLIPR instrument to each well. An 8-point concentration response curve, with final NPY concentrations of 1 nM to 3 μ M, was used to determine NPY EC_{50} .

Test compounds were dissolved in 1 μ l DMSO, diluted in 20 μ l KRH buffer with carbachol/Thomae, and added to cells prepared as described above. The 96 well plates containing prepared cells and test compounds were incubated in the dark, at room temperature for 0.5 – 6 hours. It is important that the incubation not continue beyond 6 hours. Just prior to determining the fluorescence response, 100 μ l human NPY diluted in KRH/Thomae buffer to 2 x EC_{50} was automatically added by the FLIPR instrument to each well of the 96 well plate for a final sample volume of 200 μ l and a final NPY concentration of EC_{50} . The final concentration of test compounds in the assay wells was between 1 μ M and 5 μ M. Typically cells exposed to one EC_{50} of NPY exhibit a fluorescence response of about 10,000 Relative Fluorescence Units. Antagonists of the NPY5 receptor exhibit a response that is significantly less than that of the control cells to the p 0.05 level, as measured using a parametric test of statistical significance. Typically, antagonists of the NPY5 receptor decrease the fluorescence response relative to control cells by about 20%, preferably by about 50%, and most preferably by at least 80% as compared to matched control.

- 56 -

Example 13Antagonism of Bovine Pancreatic Polypeptide-Induced Food Intake

5 This Example illustrates the use of representative compounds to decrease food intake.

Male Sprague-Dawley rats aged 7 weeks are maintained under controlled temperature ($23 \pm 3^\circ\text{C}$), humidity ($55 \pm 15\%$) and light-dark cycle (7:00-19:00 light on). Rats are housed individually with *ad libitum* access to food and water.

10 Rats are anesthetized with sodium pentobarbital (50 mg/kg, i.p.). A permanent stainless steel guide cannula for intracerebroventricular (ICV) injection (21 gauge, 10 mm long) is stereotactically implanted into the right lateral ventricle. The stereotaxic coordinates used are as follows: 0.9 mm posterior and 1.2 mm lateral to the bregma and 0.5 mm ventral to the brain surface.

15 Animals are allowed to recover at least 6 days postoperatively before the start of the feeding experiment. The day before the experiment, animals are handled and undergo mock injection, and nocturnal food intake is measured. Rats that eat more than 15 g during the night before the experiment are used for the BPP feeding antagonism experiment.

20 Test compounds are suspended in 0.5% methylcellulose and orally administered by gavage. Administration of test compounds usually begins at 10:00. Dosing volume is about 5 ml/kg. One hour after administration of the test compound, bovine pancreatic polypeptide (Bovine PP 5 μg / 10 μl / 1 min.) is injected ICV through a stainless steel injector (26 gauge) attached to a 50 μl Hamilton microsyringe by polyethylene tubing. Bovine PP is dissolved in 10 mM PBS containing 0.05% BSA.

25 Two hour post-injection food intake is measured for each rat.

Test compounds that reduce food intake two hours post-injection relative to food intake of control animals (animals inject with Bovine PP but not a test compound) are identified as compounds that antagonize bovine PP induced feeding.

Example 14

30 Food Deprivation Model

This Example illustrates the use of representative NPY5 modulators to decrease food intake within a food deprivation model.

- 57 -

Experimentally naive and experienced male Sprague-Dawley rats (Sasco, St. Louis, MO) weighing 210-300g at the beginning of the experiment are used.

Animals are triple-housed in stainless steel hanging cages in a temperature ($22^{\circ}\text{C} \pm 2^{\circ}$) and humidity (40-70% RH) controlled animal facility with a 12:12 hour light-dark cycle.

- 5 Food (Standard Rat Chow, PMI Feeds Inc., #5012) and water are available ad libitum.

Consumption data is collected while the animals were housed in Nalgene Metabolic cages (Model #650-0100). Each cage is comprised of subassemblies made of clear polymethylpentene (PMP), polycarbonate (PC), or stainless steel (SS). All parts disassemble for quick and accurate data collection and for cleaning. The entire
10 cylinder-shaped plastic and SS cage rests on a SS stand and houses one animal.

The animal is contained in the round Upper Chamber (PC) assembly (12cm high and 20cm in diameter) and rests on a SS floor. Two subassemblies are attached to the Upper Chamber. The first assembly consists of a SS feeding chamber (10cm long, 5cm high and 5cm wide) with a PC feeding drawer attached to the bottom.
15 The feeding drawer has two compartments: a food storage compartment with the capacity for approximately 50g of pulverized rat chow, and a food spillage compartment. The animal is allowed access to the pulverized chow by an opening in the SS floor of the feeding chamber. The floor of the feeding chamber does not allow access to the food dropped into the spillage compartment. The second assembly
20 includes a water bottle support, a PC water bottle (100ml capacity) and a graduated water spillage collection tube. The water bottle support funnels any spilled water into the water spillage collection tube.

The lower chamber consists of a PMP separating cone, PMP collection funnel, PMP fluid (urine) collection tube, and a PMP solid (feces) collection tube. The
25 separating cone is attached to the top of the collection funnel, which in turn is attached to the bottom of the Upper Chamber. The urine runs off the separating cone onto the walls of the collection funnel and into the urine collection tube. The separating cone also separates the feces and funnels it into the feces collection tube. Food consumption, water consumption, and body weight were measured with an Ohaus Portable Advanced
30 scale ($\pm 0.1\text{g}$ accuracy).

Prior to the day of testing, animals were habituated to the testing apparatus by placing each animal in a Metabolic cage for 1 hour. On the day of the experiment, animals that were food deprived the previous night were weighed and

- 58 -

assigned to treatment groups. Assignments were made using a quasi-random method utilizing the body weights to assure that the treatment groups had similar average body weight. Animals were then administered either vehicle (0.5% methyl cellulose, MC) or NPY5 modulator. At that time, the feeding drawer filled with pulverized chow, the filled water bottle, and the empty urine and feces collection tubes were weighed. Two hours after modulator treatment, each animal is weighed and placed in a Metabolic Cage. Following a one hour test session, animals are removed and body weight obtained. The food and water containers are then weighed and the data recorded.

NPY5 modulators (suspended in 0.5% MC) or 0.5% MC are...
administered orally (PO) using a gavage tube connected to a 3 or 5ml syringe at a volume of 10ml/kg. Each modulator was made into a homogenous suspension by stirring and ultrasonication for at least 1 hour prior to dosing.

The means and standard errors of the mean (SEM) for food consumption, water consumption, and body weight change are presented. One-way analysis of variance using Systat (5.2.1) was used to test for group differences. A significant effect is defined as having a p value of <.05.

The following parameters are defined: Body weight change is the difference between the body weight of the animal immediately prior to placement in the metabolic cage and its body weight at the end of the one hour test session. Food consumption is the difference in the weight of the food drawer prior to testing and the weight following the 1 hour test session. Water consumption is the difference in the weight of the water bottle prior to testing and the weight following the 1 hour test session. The most potent compounds of the invention significantly reduce food intake and body weight gain.

Description of the Sequence Listing

SEQ ID NO:1 is a 5' human NPY5 receptor primer
SEQ ID NO:2 is a 3' human NPY5 receptor primer
SEQ ID NO:3 is human NPY1 receptor
SEQ ID NO:4 is NPY1 amino acid sequence
SEQ ID NO:5 is the chimeric DNA sequence designated hNPY5 Y1IC3
SEQ ID NO:6 is human NPY5 receptor nucleotide sequence
SEQ ID NO:7 is human NPY5 receptor amino acid sequence
SEQ ID NO:8 is nucleotides 903-964 of human NPY5 receptor

- 59 -

SEQ ID NO:9 is amino acids 236-256 of human NPY5 receptor

SEQ ID NO:10 is the HY1L3S sense oligo

SEQ ID NO:11 is HY1L3AS' antisense oligo

SEQ ID NO:12 is the chimeric DNA sequence designated

5 hNPY5 Y1CT

SEQ ID NO:13 is primer HY1R1

SEQ ID NO:14 is primer HY5R1

SEQ ID NO:15 is forward primer corresponding to bases 527-551 of

human Y5 receptor

10 SEQ ID NO:16 is the chimeric DNA sequence designated

hNPY5 Y1IC3/ Y1CT

SEQ ID NO:17 is amino acid sequence of the chimeric receptor

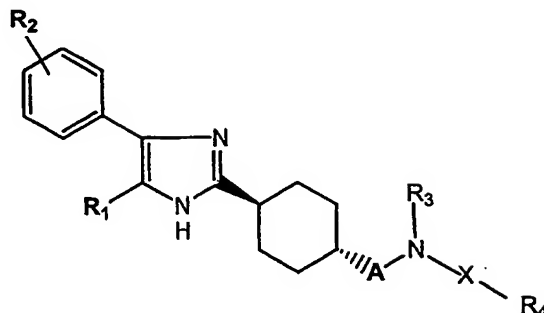
hNPY5 Y1IC3/ Y1CT

From the foregoing it will be appreciated that, although specific

15 embodiments of the invention have been described herein for purposes of illustration, various modifications may be made without deviating from the spirit and scope of the invention. Accordingly, the invention is not limited except as by the appended claims.

- 60 -
CLAIMS

1. A compound of the formula:



Formula I

- 5 or a pharmaceutically acceptable salt thereof, wherein:

R_1 :

- (i) represents hydrogen, C_1 - C_8 alkyl, C_2 - C_8 alkenyl, C_2 - C_8 alkynyl, C_1 - C_8 alkoxy, C_1 - C_8 alkanoyloxy, C_1 - C_8 carbonate, C_1 - C_8 carbamate, C_1 - C_8 alkylthio, halo(C_1 - C_8)alkyl, halo(C_1 - C_8)alkoxy, hydroxy(C_1 - C_8)alkyl, hydroxy(C_1 - C_8)alkoxy, mono or di(C_1 - C_8)alkyl amino, C_1 - C_8 alkanoyl, C_1 - C_8 alkoxycarbonyl, -
10 $COOH$, $-SO_2NH_2$, mono or dialkylsulfonamido, $-C(O)NH_2$ or mono or di(C_1 - C_8)alkylcarboxamido; or

(ii) taken together with R_2 , represents a 5- to 7-member carbocyclic or heterocyclic ring;

- 15 R_2 represents 0 to 5 ring substituents, each substituent independently selected from hydrogen, halogen, hydroxyl, amino, cyano, nitro, C_1 - C_8 alkyl, C_2 - C_8 alkenyl, C_2 - C_8 alkynyl, C_1 - C_8 alkoxy, C_1 - C_8 alkanoyloxy, C_1 - C_8 carbonate, C_1 - C_8 carbamate, C_1 - C_8 alkylthio, halo(C_1 - C_8)alkyl, halo(C_1 - C_8)alkoxy, hydroxy(C_1 - C_8)alkyl, hydroxy(C_1 - C_8)alkoxy, mono or di(C_1 - C_8)alkyl amino, C_1 - C_8 alkanoyl, C_1 - C_8
20 alkoxycarbonyl, $-COOH$, $-SO_2NH_2$, mono and dialkylsulfonamido, $-C(O)NH_2$, mono and di(C_1 - C_8) alkylcarboxamido, and substituents that, taken together with R_1 , form a 5- to 7-member carbocyclic or heterocyclic ring;

A represents $-CO-$ or $-(CH_2)_n-$, wherein n is an integer ranging from 1 to 3;

- 25 R_3 represents:

(i) hydrogen;

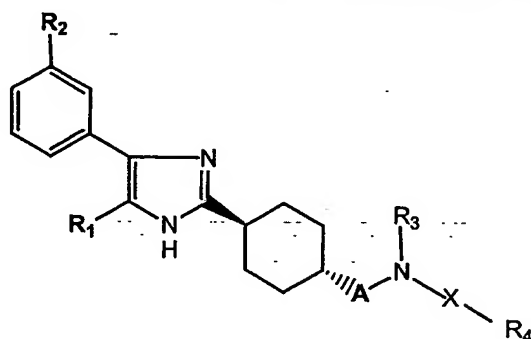
- 61 -

- (ii) C₁-C₈ alkyl, optionally substituted with from 1 to 8 substituents that are independently selected from halogen, hydroxyl, carbocyclic groups and heterocyclic groups, wherein each carbocyclic or heterocyclic group contains from 3 to 10 ring members; or
- 5 (iii) a bond to R₄, forming a heterocyclic group having from 1 to 3 fused or pendant rings, each ring containing from 5 to 10 ring members, wherein each ring is optionally substituted by from 1 to 5 substituents that are independently selected from halogen, cyano, nitro, C₁-C₈ alkyl, C₂-C₈ alkenyl, C₂-C₈ alkynyl, C₁-C₈ alkoxy, C₁-C₈ alkylthio, hydroxy, amino, mono or di(C₁-C₈)alkyl amino, halo(C₁-C₈)alkyl, halo(C₁-C₈)alkoxy, hydroxy(C₁-C₈)alkyl, hydroxy(C₁-C₈)alkoxy C₂-C₈ alkanoyl, C₁-C₈ alkoxycarbonyl, -COOH, -SO₂NH₂, mono or dialkylsulfonamido, -C(O)NH₂ and mono and di(C₁-C₈)alkylcarboxamido;
- X represents a bond, -S(O)₂-, -C(O)- or -NHC(O)-; and
- R₄:
- 15 (i) represents hydrogen;
- (ii) represents C₁-C₈ alkyl, C₂-C₈ alkenyl, C₂-C₈ alkynyl, C₁-C₈ alkoxy, C₁-C₈ alkanoyloxy, C₁-C₈ carbonate, halo(C₁-C₈)alkyl, halo(C₁-C₈)alkoxy, hydroxy(C₁-C₈)alkyl, hydroxy(C₁-C₈)alkoxy, C₁-C₈ alkanone or C₁-C₈ alkyl ether, optionally substituted with from 1 to five substituents that are independently selected
- 20 from halogen, hydroxyl, carbocyclic groups and heterocyclic groups, wherein each carbocyclic or heterocyclic group contains from 5 to 10 ring members;
- (iii) represents a carbocyclic or heterocyclic group having from 1 to 3 fused or pendant rings, each ring containing from 5 to 10 ring members, wherein each ring is optionally substituted by from 1 to 5 substituents that are
- 25 independently selected from halogen, cyano, nitro, C₁-C₈ alkyl, C₂-C₈ alkenyl, C₂-C₈ alkynyl, C₁-C₈ alkoxy, C₁-C₈ alkanoyloxy, C₁-C₈ carbonate, C₁-C₈ carbamate, C₁-C₈ alkylthio, hydroxy, amino, mono or di(C₁-C₈)alkyl amino, halo(C₁-C₈)alkyl, halo(C₁-C₈)alkoxy, hydroxy(C₁-C₈)alkyl, hydroxy(C₁-C₈)alkoxy C₂-C₈ alkanoyl, C₁-C₈ alkoxycarbonyl, -COOH, -SO₂NH₂, mono or dialkylsulfonamido, -C(O)NH₂ and mono
- 30 or di(C₁-C₈)alkylcarboxamido; or
- (iv) taken together with R₃ and X, represents a heterocyclic group having from 1 to 3 fused or pendant rings, each ring containing from 5 to 10 ring members, wherein each ring is optionally substituted by from 1 to 5 substituents that are

- 62 -

independently selected from halogen, cyano, nitro, C₁-C₈ alkyl, C₂-C₈ alkenyl, C₂-C₈ alkynyl, C₁-C₈ alkoxy, C₁-C₈ alkanoyloxy, C₁-C₈ carbonate, C₁-C₈ carbamate, C₁-C₈ alkylthio, hydroxy, amino, mono or di(C₁-C₈)alkyl amino, halo(C₁-C₈)alkyl, halo(C₁-C₈)alkoxy, hydroxy(C₁-C₈)alkyl, hydroxy(C₁-C₈)alkoxy C₂-C₈ alkanoyl, C₁-C₈ alkoxycarbonyl, COOH, -SO₂NH₂, mono or dialkylsulfonamido, -C(O)NH₂ and mono or di(C₁-C₈)alkylcarboxamido.

2. A compound according to claim 1, wherein the compound further satisfies Formula II, and wherein R₂ is trifluoromethyl, halogen or cyano.



Formula II

3. A compound according to claim 1, wherein R₃ is hydrogen, methyl, ethyl or propyl.

4. A compound according to claim 1, wherein R₄ is:

(i) an aromatic group selected from phenyl, benzyl, phenoxy, benzoxyl, phenylethanonyl, pyrimidin-2-yl, pyrimidin-4-yl, pyrimidin-5-yl, tetrahydropyran-2-yl, indan-1-yl, tetrazolyl, pyridin-2-yl, pyridin-3-yl and pyridin-4-yl, wherein the group is optionally substituted by from 1 to 3 substituents independently selected from hydroxyl, halogen and C₁-C₆ alkyl;

(ii) a non-aromatic group selected from hydrogen, C₁-C₆ alkyl, C₁-C₆ alkoxy, halo(C₁-C₆)alkyl, halo(C₁-C₆)alkoxy, hydroxy(C₁-C₆)alkyl, hydroxy(C₁-C₆)alkoxy, C₁-C₆ alkanone or C₁-C₆ alkyl ether; or

(iii) taken with R₃ and X to form a heterocyclic group selected from tetrazolyl, morpholin-4-yl, pyrimidin-2-yl, pyridin-2-yl, pyridin-3-yl and pyridin-4-yl, piperidin-1-yl, pyrrolidin-1-yl, each of which is optionally substituted with from 1 to 3 substituents independently selected from halogen, C₁-C₆ alkyl, C₁-C₆ alkoxy, hydroxyl, halo(C₁-C₆)alkyl and halo(C₁-C₆)alkoxy.

5. A compound according to claim 1, wherein A is -CH₂-.

- 63 -

6. A compound according to claim 1, wherein X is $-\text{S}(\text{O})_2-$, R_3 is hydrogen and R_4 is a group selected from phenyl, pyridyl and pyrimidyl, wherein the group is optionally substituted by from 1 to 3 substituents independently selected from hydroxyl, halogen and C_1 - C_6 alkyl.
- 5 7. A compound according to claim 1, wherein A is $-\text{CH}_2-$, X is $-\text{C}(\text{O})-$, R_3 is hydrogen and R_4 is C_1 - C_6 alkyl, C_1 - C_6 alkoxy, halo(C_1 - C_6)alkyl or halo(C_1 - C_6)alkoxy.
8. A compound according to claim 1, wherein A is $-\text{C}(\text{O})-$, X is $-\text{NHC}(\text{O})-$, R_3 is hydrogen and R_4 is C_1 - C_6 alkyl.
- 10 9. A compound according to claim 1, wherein A is $-\text{C}(\text{O})-$, X is a bond, R_3 is hydrogen or C_1 - C_6 alkyl, and R_4 is C_1 - C_6 alkyl, halo(C_1 - C_6)alkyl, hydroxy(C_1 - C_6)alkyl, or an aromatic group selected from phenyl, pyrimidin-2-yl, indan-1-yl, wherein the group is optionally substituted by from 1 to 3 substituents independently selected from hydroxyl, halogen, C_1 - C_6 alkyl, hydroxy(C_1 - C_6)alkyl and C_1 - C_6 alkanone.
- 15 10. A compound according to claim 1, wherein the compound is selected from:
 - (a) 2-Chloro-N-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]cyclohexanecarbonyl}-benzene sulfonamide;
 - 20 (b) 4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid(2-hydroxy-propyl)-methyl-amide;
 - (c) 4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid(2-oxo-2-phenyl-ethyl)-amide;
 - (d) Pyridine-3-sulfonic acid{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-amide;
 - 25 (e) 4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid(2-hydroxy-2-phenyl-ethyl)-amide;
 - (f) 1-{4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-1H-tetrazole;
 - 30 (g) N-{4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-benzenesulfonamide;
 - (h) 4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid(2-hydroxy-indan-1-yl)-amide;

- 64 -

- (i) {4-[4-(3-Chloro-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-carbamic acid ethyl ester;
- (j) Cyclopentyl-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-amine;
- 5 (k) N-{4-[4-(4-Bromo-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-2,2,2-trifluoro-acetamide;
- (l) N-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarbonyl}-benzene sulfonamide;
- (m) Pyrimidin-2-yl-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-amine;
- 10 (n) 4-[4-(3-Bromo-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid N-acetyl-hydrazide;
- (o) 4,N-Dimethyl-N-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarbonyl}-benzene sulfonamide;
- 15 (p) N-{4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-acetamide;
- (q) Benzenesulfinic acid {4-[4-(3-chloro-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-amide;
- (r) 4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid(2-hydroxy-1,1-dimethyl-ethyl)-amide;
- 20 (s) 4-Hydroxy-N-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-butyramide;
- (t) 4-methyl-N-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarbonyl}-benzene sulfonamide;
- 25 (u) N-{4-[4-(3-Bromo-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-trifluoroacetamide;
- (v) 4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid (2-hydroxy-propyl)-amide;
- (w) N-{4-[5-methyl-4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-acetamide;
- 30 (x) 2,2,2-Trifluoro-N-{4-[4-(4-methoxy-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-acetamide;
- (y) 4-Chloro-N-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-

- 65 -

cyclohexanecarbonyl}-benzene sulfonamide;

(z) N-[4-(8-Methoxy-4,5-dihydro-3H-naphtho[1,2-d]imidazol-2-yl)-cyclohexylmethyl]-benzenesulfonamide;

(aa) 4-[4-(3-Bromo-phenyl)-1H-imidazol-2-yl]-
5 cyclohexanecarboxylic acid pyrimidin-2-ylamide;

(bb) N-Methyl-N-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-benzenesulfonamide;

(cc) N-{4-[4-(3-Bromo-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-acetamide;

10 (dd) 5-Methyl-pyridine-2-sulfonic acid {4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarbonyl}-amide;

(ee) 4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid (1-hydroxymethyl-2-methyl-propyl)-amide;

(ff) {4-[5-Methyl-4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-
15 cyclohexylmethyl}-carbamic acid benzyl ester;

(gg) [4-(8-Methoxy-4,5-dihydro-3H-naphtho[1,2-d]imidazol-2-yl)-cyclohexylmethyl]-carbamic acid benzyl ester;

(hh) 4-[4-(3-Trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexanecarboxylic acid N-acetyl-hydrazide;

20 (ii) 2,2,2-Trifluoro-N-[4-(4-phenyl-1H-imidazol-2-yl)-cyclohexylmethyl]-acetamide; and

(jj) 2,2,2-Trifluoro-N-{4-[4-(3-trifluoromethyl-phenyl)-1H-imidazol-2-yl]-cyclohexylmethyl}-acetamide.

11. A compound according to claim 1, wherein the compound exhibits a K_i
25 of 1 micromolar or less in an NPY5 receptor ligand binding assay.

12. A compound according to claim 1, wherein the compound exhibits a K_i
of 100 nanomolar or less in an NPY5 receptor ligand binding assay.

13. A compound according to claim 1, wherein the compound exhibits a K_i
of 10 nanomolar or less in an NPY5 receptor ligand binding assay.

30 14. An NPY5 receptor modulator, comprising a compound according to claim 1.

- 66 -

15. A pharmaceutical composition comprising a compound according to claim 1 or a modulator according to claim 14 in combination with a physiologically acceptable carrier or excipient.
16. A pharmaceutical composition according to claim 15, wherein the
5 composition is formulated as an injectible fluid, an aerosol, a cream, a gel, a pill, a capsule, a syrup or a transdermal patch.
17. A method for treating a disease or disorder associated with NPY5 receptor activation, comprising administering to a patient in need of such treatment an effective amount of a compound according to claim 1 or a modulator according to claim
10 14.
18. A method according to claim 17 wherein the disease or disorder is an eating disorder, a psychiatric disorder, a cardiovascular disorder or diabetes.
19. A method according to claim 17, wherein the compound or modulator is administered orally.
- 15 20. A method according to claim 17, wherein the compound or modulator is administered intranasally, intravenously or topically.
21. A method according to claim 17, wherein the patient is a human.
22. A compound according to claim 1, wherein the compound is radiolabeled.
- 20 23. A method for determining the presence or absence of NPY5 receptor in a sample, comprising the steps of:
- (a) contacting a sample with an agent comprising a compound according to claim 1 under conditions that permit binding of the agent to NPY5 receptor; and
- 25 (b) detecting a level of agent bound to NPY5 receptor, and therefrom determining the presence or absence of NPY-5 receptor in the sample.
24. A method according to claim 23, wherein the agent is a radiolabeled compound according to claim 22, and wherein the step of detection comprises the steps of:
- 30 (i) separating unbound agent from bound agent; and
(ii) detecting the presence or absence of bound agent in the sample.
25. A method according to claim 23, wherein the step of detection is accomplished using radiography.

- 67 -

26. A method for modulating binding of NPY to NPY5 receptor *in vitro*, the method comprising contacting NPY5 receptor with a compound according to claim 1 or a modulator according to claim 14 under conditions and in an amount sufficient to detectably modulate NPY binding to NPY5 receptor.
- 5 27. A method for modulating binding of NPY to NPY5 receptor, the method comprising contacting cells expressing NPY5 receptor with a compound according to claim 1 or a modulator according to claim 14 in an amount sufficient to detectably modulate NPY binding to cells expressing a cloned NPY5 receptor *in vitro*.
28. A method for modulating binding of NPY to NPY5 receptor in an
10 patient, comprising administering to a patient a compound according to claim 1 or a modulator according to claim 14, and thereby modulating binding of NPY to NPY5 receptor in the patient.
29. A method according to any one of claims 26-28, wherein the modulation is inhibition.
- 15 30. A method according to any one of claims 26-28, wherein the NPY5 receptor is a human NPY5 receptor.
31. A packaged pharmaceutical preparation, comprising:
(a) a pharmaceutical composition according to claim 15 in a
container;
20 and
(b) instructions for using the composition to treat a patient suffering from a disorder responsive to NPY-5 receptor antagonism or agonism.
32. A packaged pharmaceutical preparation according to claim 31, wherein the patient is suffering from an eating disorder, a psychiatric disorder, a cardiovascular
25 disorder or diabetes.---

SEQUENCE LISTING

<110> Blum, Charles
Brielmann, Harry
De Lombaert, Stephane
Zheng, Xiaozhang

<120> SUBSTITUTE 2-CYCLOHEXYL-4-PHENYL-1H-IMIDAZOLE
DERIVATIVES

<130> N01.2000P

<140> N/A

<141> 2002-10-21

<160> 17

<170> PatentIn Ver. 2.0

<210> 1

<211> 21

<212> DNA

<213> Homo sapiens

<400> 1

ttttggttgc tgacaaatgt c

21

<210> 2

<211> 26

<212> DNA

<213> Homo sapiens

<400> 2

ccttggtaaa cagtgagaat tattac

26

<210> 3

<211> 1605

<212> DNA

<213> Homo sapiens

<400> 3

ccttcttttaa tgaagcagga gcgaaaaaga caaattccaa agaggattgt tcagttcaag 60
ggaatgaaga attcagaata attttggttaa atggattcca atatggggaa taagaataag 120
ctgaacagtt gacctgcttt gaagaaacat actgtccatt tgtctaaaat aatctataac 180
aaccaaacca atcaaaatga attcaacatt attttcccag gttgaaaatc attcagtcca 240
ctctaatttc tcagagaaga atgccagct tctggctttt gaaaatgatg attgtcatct 300
gcccttggtc atgatatta ccttagctct tgcttatgga gctgtgatca ttcttggtgt 360
ctctggaaac ctggccttga tcataatcat cttgaaacaa aaggagatga gaaatgttac 420

```

caacatcctg attgtgaacc tttccttctc agacttgctt gttgccatca tgtgtctccc 480
ctttacattt gtctacacat taatggacca ctgggtcttt ggtgaggcga tgtgtaagtt 540
gaatcctttt gtgcaatgtg tttcaatcac tgtgtccatt ttctctctgg ttctcattgc 600
tgtggaacga catcagctga taatcaaccc tcgagggtgg agaccaaata atagacatgc 660
ttatgtaggt attgctgtga tttgggtcct tgctgtggct tcttctttgc ctttcttgat 720
ctaccaagta atgactgatg agccgttcca aaatgtaaca cttgatgcgt acaaagacaa 780
atacgtgtgc tttgatcaat ttccatcgga ctctcatagg ttgtcttata ccactctcct 840
cttggtgctg cagtattttg gtccactttg ttttatattt atttgctact tcaagatata 900
tatacgctta aaaaggagaa acaacatgat ggacaagatg agagacaata agtacaggtc 960
cagtgaaccc aaaagaatca atatcatgct gctctccatt gtggtagcat ttgcagctcg 1020
ctggctccct cttaccatct ttaacactgt gtttgattgg aatcatcaga tcattgctac 1080
ctgcaaccac aatctgttat tcctgctctg ccacctcaca gcaatgatat ccactgtgtg 1140
caaccccata ttttatgggt tcctgaacaa aaacttccag agagacttgc agttcttctt 1200
caacttttgt gatttccggg ctccggatga tgattatgaa acaatagcca tgtccacgat 1260
gcacacagat gtttccaaaa cttctttgaa gcaagcaagc ccagtcgcat ttaaaaaaat 1320
caacaacaat gatgataatg aaaaaatctg aaactactta tagcctatgg tcccgatga 1380
catctgttta aaaacaagca caacctgcaa catactttga ttacctgttc tcccaaggaa 1440
tggggttgaa atcatttgaa aatgactaag atttcttgtt cttgcttttt actgcttttg 1500
ttgtagttgt cataattaca tttggaacaa aagggtgtgg ctttggggtc ttctggaaat 1560
agttttgacc agacatcttt gaagtgtctt ttgtgaattt accag 1605

```

<210> 4

<211> 384

<212> PRT

<213> Homo sapiens

<400> 4

```

Met Asn Ser Thr Leu Phe Ser Gln Val Glu Asn His Ser Val His Ser
1           5           10           15

```

```

Asn Phe Ser Glu Lys Asn Ala Gln Leu Leu Ala Phe Glu Asn Asp Asp
20           25           30

```

```

Cys His Leu Pro Leu Ala Met Ile Phe Thr Leu Ala Leu Ala Tyr Gly
35           40           45

```

```

Ala Val Ile Ile Leu Gly Val Ser Gly Asn Leu Ala Leu Ile Ile Ile
50           55           60

```

```

Ile Leu Lys Gln Lys Glu Met Arg Asn Val Thr Asn Ile Leu Ile Val
65           70           75           80

```

```

Asn Leu Ser Phe Ser Asp Leu Leu Val Ala Ile Met Cys Leu Pro Phe
85           90           95

```

```

Thr Phe Val Tyr Thr Leu Met Asp His Trp Val Phe Gly Glu Asn Met
100          105          110

```

Cys	Lys	Leu	Asn	Pro	Phe	Val	Gln	Cys	Val	Ser	Ile	Thr	Val	Ser	Ile	115	120	125	
Phe	Ser	Leu	Val	Leu	Ile	Ala	Val	Glu	Arg	His	Gln	Leu	Ile	Ile	Asn	130	135	140	
Pro	Arg	Gly	Trp	Arg	Pro	Asn	Asn	Arg	His	Ala	Tyr	Val	Gly	Ile	Ala	145	150	155	160
Val	Ile	Trp	Val	Leu	Ala	Val	Ala	Ser	Ser	Leu	Pro	Phe	Leu	Ile	Tyr	165	170	175	
Gln	Val	Met	Thr	Asp	Glu	Pro	Phe	Gln	Asn	Val	Thr	Leu	Asp	Ala	Tyr	180	185	190	
Lys	Asp	Lys	Tyr	Val	Cys	Phe	Asp	Gln	Phe	Pro	Ser	Asp	Ser	His	Arg	195	200	205	
Leu	Ser	Tyr	Thr	Thr	Leu	Leu	Leu	Val	Leu	Gln	Tyr	Phe	Gly	Pro	Leu	210	215	220	
Cys	Phe	Ile	Phe	Ile	Cys	Tyr	Phe	Lys	Ile	Tyr	Ile	Arg	Leu	Lys	Arg	225	230	235	240
Arg	Asn	Asn	Met	Met	Asp	Lys	Met	Arg	Asp	Asn	Lys	Tyr	Arg	Ser	Ser	245	250	255	
Glu	Thr	Lys	Arg	Ile	Asn	Ile	Met	Leu	Leu	Ser	Ile	Val	Val	Ala	Phe	260	265	270	
Ala	Val	Cys	Trp	Leu	Pro	Leu	Thr	Ile	Phe	Asn	Thr	Val	Phe	Asp	Trp	275	280	285	
Asn	His	Gln	Ile	Ile	Ala	Thr	Cys	Asn	His	Asn	Leu	Leu	Phe	Leu	Leu	290	295	300	
Cys	Ala	Leu	Thr	Ala	Met	Ile	Ser	Thr	Cys	Val	Asn	Pro	Ile	Phe	Tyr	305	310	315	320
Gly	Phe	Leu	Asn	Lys	Asn	Phe	Gln	Arg	Asp	Ser	Gln	Phe	Phe	Phe	Asn	325	330	335	
Phe	Cys	Asp	Phe	Arg	Ser	Arg	Asp	Asp	Asp	Tyr	Glu	Thr	Ile	Ala	Met	340	345	350	
Ser	Thr	Met	His	Thr	Asp	Val	Ser	Lys	Thr	Ser	Leu	Lys	Gln	Ala	Ser	355	360	365	

Pro Val Ala Phe Lys Lys Ile Asn Asn Asn Asp Asp Asn Glu Lys Ile
 370 375 380

<210> 5
 <211> 1069
 <212> DNA
 <213> Homo sapiens

<400> 5
 ttttggttgc tgacaaatgt ctttttattc caagcaggac tataatatgg atttagagct 60
 cgacgagtat tataacaaga cacttgccac agagaataat actgctgccca ctcggaattc 120
 tgatttccca gtctgggatg actataaaag cagtgtagat gacttacagt attttctgat 180
 tgggctctat acatttgtaa gtcttcttgg ctttatgggg aatctactta ttttaatggc 240
 tctcatgaaa aagcgtaatc agaagactac ggtaaacttc ctcataggca atctggcctt 300
 ttctgatatc ttggttggtc tgttttgctc acctttcaca ctgacgtctg tcttgctgga 360
 tcagtggatg tttggcaaag tcatgtgccca tattatgcct tttcttcaat gtgtgtcagt 420
 tttggtttca actttaattt taatatcaat tgccattgtc aggtatcata tgataaaaca 480
 tcccatatct aataatttaa cagcaaacca tggctacttt ctgatatgcta ctgtctggac 540
 actagggtttt gccatctggt ctcccccttc agtggtttcac agtcttgtgg aacttcaaga 600
 aacatttgggt tcagcattgc tgagcagcag gtatttatgt gttgagtcac ggccatctga 660
 ttcatacaga attgccttta ctatctcttt attgctagtt cagtatattc tgcccttagt 720
 ttgtcttact gtaagtcata caagtgtctg catacgccta aaaaggagaa acaacatgat 780
 ggacaagatg agagacaata agtacaggtc cagtagatct cgaagtgttt tctacagact 840
 gaccatactg atattagtat ttgctgttag ttggatgccca ctacaccttt tccatgtggg 900
 aactgatttt aatgacaatc ttatttcaaa taggcatttc aagttgggtg attgcatttg 960
 tcatttggtg ggcattgatg cctgttgtct taatccaatt ctatatgggt ttcttaataa 1020
 tgggattaaa gctgatttag tgtcccttat acactgtctt catatgtaa 1069

<210> 6
 <211> 1406
 <212> DNA
 <213> Homo sapiens

<400> 6
 ttttggttgc tgacaaatgt ctttttattc caagcaggac tataatatgg atttagagct 60
 cgacgagtat tataacaaga cacttgccac agagaataat actgctgccca ctcggaattc 120
 tgatttccca gtctgggatg actataaaag cagtgtagat gacttacagt attttctgat 180
 tgggctctat acatttgtaa gtcttcttgg ctttatgggg aatctactta ttttaatggc 240
 tctcatgaaa aagcgtaatc agaagactac ggtaaacttc ctcataggca atctggcctt 300
 ttctgatatc ttggttggtc tgttttgctc acctttcaca ctgacgtctg tcttgctgga 360
 tcagtggatg tttggcaaag tcatgtgccca tattatgcct tttcttcaat gtgtgtcagt 420
 tttggtttca actttaattt taatatcaat tgccattgtc aggtatcata tgataaaaca 480
 tcccatatct aataatttaa cagcaaacca tggctacttt ctgatatgcta ctgtctggac 540
 actagggtttt gccatctggt ctcccccttc agtggtttcac agtcttgtgg aacttcaaga 600


```

aacatttggg tcagcattgc tgagcagcag gtatttatgt gttgagtcac ggccatctga 660
ttcatacaga attgccttta ctatctcttt attgctagtt cagtatatcc tgcccttagt 720
ttgtcttact gtaagtcata caagtgtctg cagaagtata agctgtggat tgtccaacaa 780
agaaaacaga cttgaagaaa atgagatgat caacttaact cttcatccat ccaaaaagag 840
tgggcctcag gtgaaactct ctggcagcca taaatggagt tattcattca tcaaaaaaca 900
cagaagaaga tatagcaaga agacagcatg tgtgttacct gctccagaaa gaccttctca 960
agagaaccac tccagaatac ttccagaaaa ctttggctct gtaagaagtc agctctcttc 1020
atccagtaag ttcataccag gggccccac ttgctttgag ataaaacctg aagaaaattc 1080
agatgttcat gaattgagag taaaacgttc tgttacaaga ataaaaaaga gatctcgaag 1140
tgttttctac agactgacca tactgatatt agtatttgct gttagttgga tgccactaca 1200
ccttttccat gtggttaactg attttaatga caatcttatt tcaaataggc atttcaagtt 1260
ggtgtattgc atttgtcatt tgttgggcat gatgtcctgt tgtcttaatc caattctata 1320
tggttttctt aataatggga ttaaagctga tttagtgtcc cttatacact gtcttcatat 1380
gtaataattc tcactgttta ccaagg                                     1406

```

<210> 7

<211> 455

<212> PRT

<213> Homo sapiens

<400> 7

```

Met Ser Phe Tyr Ser Lys Gln Asp Tyr Asn Met Asp Leu Glu Leu Asp
  1                      5                      10                      15

```

```

Glu Tyr Tyr Asn Lys Thr Leu Ala Thr Glu Asn Asn Thr Ala Ala Thr
      20                      25                      30

```

```

Arg Asn Ser Asp Phe Pro Val Trp Asp Asp Tyr Lys Ser Ser Val Asp
      35                      40                      45

```

```

Asp Leu Gln Tyr Phe Leu Ile Gly Leu Tyr Thr Phe Val Ser Leu Leu
      50                      55                      60

```

```

Gly Phe Met Gly Asn Leu Leu Ile Leu Met Ala Leu Met Lys Lys Arg
      65                      70                      75                      80

```

```

Asn Gln Lys Thr Thr Val Asn Phe Leu Ile Gly Asn Leu Ala Phe Ser
      85                      90                      95

```

```

Asp Ile Leu Val Val Leu Phe Cys Ser Pro Phe Thr Leu Thr Ser Val
      100                      105                      110

```

```

Leu Leu Asp Gln Trp Met Phe Gly Lys Val Met Cys His Ile Met Pro
      115                      120                      125

```

```

Phe Leu Gln Cys Val Ser Val Leu Val Ser Thr Leu Ile Leu Ile Ser
      130                      135                      140

```

Ile Ala Ile Val Arg Tyr His Met Ile Lys His Pro Ile Ser Asn Asn
 145 150 155 160
 Leu Thr Ala Asn His Gly Tyr Phe Leu Ile Ala Thr Val Trp Thr Leu
 165 170 175
 Gly Phe Ala Ile Cys Ser Pro Leu Pro Val Phe His Ser Leu Val Glu
 180 185 190
 Leu Gln Glu Thr Phe Gly Ser Ala Leu Leu Ser Ser Arg Tyr Leu Cys
 195 200 205
 Val Glu Ser Trp Pro Ser Asp Ser Tyr Arg Ile Ala Phe Thr Ile Ser
 210 215 220
 Leu Leu Leu Val Gln Tyr Ile Leu Pro Leu Val Cys Leu Thr Val Ser
 225 230 235 240
 His Thr Ser Val Cys Arg Ser Ile Ser Cys Gly Leu Ser Trp Lys Glu
 245 250 255
 Asn Arg Leu Glu Glu Asn Glu Met Ile Asn Leu Thr Leu His Pro Ser
 260 265 270
 Lys Lys Ser Gly Pro Gln Val Lys Leu Ser Gly Ser His Lys Trp Ser
 275 280 285
 Tyr Ser Phe Ile Lys Lys His Arg Arg Arg Tyr Ser Lys Lys Thr Ala
 290 295 300
 Cys Val Leu Pro Ala Pro Glu Arg Pro Ser Gln Glu Asn His Ser Arg
 305 310 315 320
 Ile Leu Pro Glu Asn Phe Gly Ser Val Arg Ser Gln Leu Ser Ser Ser
 325 330 335
 Ser Lys Phe Ile Pro Gly Val Pro Thr Cys Phe Glu Ile Leu Pro Glu
 340 345 350
 Glu Asn Ser Asp Val His Glu Leu Arg Val Lys Arg Ser Val Thr Arg
 355 360 365
 Ile Lys Lys Arg Ser Arg Ser Val Phe Tyr Arg Leu Thr Ile Leu Ile
 370 375 380
 Leu Val Phe Ala Val Ser Trp Met Pro Leu His Leu Phe His Val Val
 385 390 395 400

Thr Asp Phe Asn Asp Asn Leu Ile Ser Asn Arg His Phe Lys Leu Val
 405 410 415

Tyr Cys Ile Cys His Leu Leu Gly Met Met Ser Cys Cys Leu Asn Pro
 420 425 430

Ile Leu Tyr Gly Phe Leu Asn Asn Gly Ile Lys Ala Asp Leu Val Ser
 435 440 445

Leu Ile His Cys Leu His Met
 450 455

<210> 8

<211> 62

<212> DNA

<213> Homo sapiens

<400> 8

tacgcctaaa aaggagaaac aacatgatgg acaagatgag agacaataag tacagggtcca 60
 gt 62

<210> 9

<211> 21

<212> PRT

<213> Homo sapiens

<400> 9

Ile Arg Leu Lys Arg Arg Asn Asn Met Met Asp Lys Met Arg Asp Asn
 1 5 10 15

Lys Tyr Arg Ser Ser
 20

<210> 10

<211> 63

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: CHIMERIC Y1/Y5
 PRIMER

<400> 10

tacgcctaaa aaggagaaac aacatgatgg acaagatgag agacaataag tacagggtcca 60
 gta 63

<210> 11

<211> 71

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: CHIMERIC Y1/Y5
PRIMER

<400> 11

```

gatctactgg acctgtactt attgtctctc atcttgtcca tcatgttggt tctccttttt 60
aggcgtatgc a

```

71

<210> 12

<211> 1500

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Y5/Y1 CHIMERA

<400> 12

```

atgtcttttt attccaagca ggactataat atggatttag agctcgacga gtattataac 60
aagacacttg ccacagagaa taatactgct gccactcgga attctgattt cccagtctgg 120
gatgactata aaagcagtgt agatgactta cagtattttc tgattgggct ctatacattt 180
gtaagtcttc ttggctttat ggggaatcta cttattttaa tggctctcat gaaaaagcgt 240
aatcagaaga ctacggtaaa cttcctcata ggcaatctgg ctttttctga tatcttggtt 300
gtgctgtttt getcaccttt cacaotgacg tctgtcttgc tggatcagtg gatgtttggc 360
aaagtcatgt gccatattat gccttttctt caatgtgtgt cagttttggg ttcaacttta 420
attttaatat caattgccat tgtcagggtat catatgataa aacatcccat atctaataat 480
ttaacagcaa accatggcta ctttctgata gctactgtct ggacactagg ttttgccatc 540
tgttctcccc ttccagtgtt tcacagtctt gtggaacttc aagaaacatt tggttcagca 600
ttgctgagca gcaggatatt atgtgttgag tcatggccat ctgattcata cagaattgcc 660
tttactatct ctttattgct agttcagtat attctgccct tagtttgtct tactgtaagt 720
catacaagtg tctgcagaag tataagctgt ggattgtcca acaaagaaa cagacttgaa 780
gaaaatgaga tgatcaactt aactcttcat ccatccaaaa agagtgggcc tcagggtgaa 840
ctctctggca gccataaatg gagttattca ttcataaaaa aacacagaag aagatatagc 900
aagaagacag catgtgtgtt acctgctcca gaaagacctt ctcaagagaa ccactccaga 960
atacttccag aaaacttttg ctctgtaaga agtcagctct cttcatccag taagttcata 1020
ccagggggtcc ccacttgctt tgagataaaa cctgaagaaa attcagatgt tcatgaattg 1080
agagtaaaac gttctgttac aagaataaaa aagagatctc gaagtgtttt ctacagactg 1140
accatactga tattagtatt tgctgttagt tggatgccac tacacctttt ccatgtggtt 1200
actgatttta atgacaatct tatttcaaat aggcatttca agttgggtgta ttgcatttgt 1260
catttggttg gcatgatgtc ctgttgtctt aatccaattc tatatgggtt tcttaataat 1320
ggaattcaga gagacttgca gttcttcttc aacttttgtg atttccggtc tcgggatgat 1380
gattatgaaa caatagccat gtccacgatg cacacagatg tttccaaaac ttctttgaag 1440
caagcaagcc cagtcgcatt taaaaaatc aacaacaatg atgataatga aaaaatctga 1500

```

<210> 13

<211> 31

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: MUTAGENIC R1
PRIMER

<400> 13

gaacaaaaga attcagagag acttgagtt c

31

<210> 14

<211> 28

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: MUTAGENIC R1
PRIMER

<400> 14

cagcttgaat tccattatta agaaaccc

28

<210> 15

<211> 25

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: PRIMER

<400> 15

gctactgtct ggacactagg ttttg

25

<210> 16

<211> 1201

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Y5/Y1 CHIMERA

<400> 16

ttttggttgc tgacaaatgt ctttttattc caagcaggac tataatatgg atttagagct 60
 cgacgagtat tataacaaga cacttgccac agagaataat actgctgcca ctcggaattc 120
 tgatttccca gtctgggatg actataaaag cagtgtagat gacttacagt attttctgat 180
 tgggctctat acatttgtaa gtcttcttgg ctttatgggg aatctactta ttttaatggc 240

```

tctcatgaaa aagcgtaatc agaagactac ggtaaacttc ctcataggca atctggcctt 300
ttctgataac ttggttggtg tgttttgctc acctttcaca ctgacgtctg tottgctgga 360
tcagtggatg tttggcaaag tcatgtgcca tattatgcct tttcttcaat gtgtgtcagt 420
tttggtttca actttaattt taatatcaat tgccattgtc aggtatcata tgataaaaca 480
tcccatatct aataatttaa cagcaaacca tggctacttt ctgataagcta ctgtctggac 540
actaggtttt gccatctgtt ctcccccttc agtgtttcac agtcttgtgg aacttcaaga 600
aacatttggt tcagcattgc tgagcagcag gtatttatgt gttgagtcac ggccatctga 660
ttcatacaga attgccttta ctatctcttt attgctagtt cagtatatc tgcccttagt 720
ttgtcttact gtaagtcata caagtgtctg catacgccta aaaaggagaa acaacatgat 780
ggacaagatg agagacaata agtacaggtc cagtagatct cgaagtgttt tctacagact 840
gaccatactg atattagtat ttgctgttag ttggatgcca ctacaccttt tccatgtggt 900
aactgatttt aatgacaatc ttatttcaaa taggcatttc aagttggtgt attgcatttg 960
tcatttggtg ggcattgatg cctgttgtct taatccaatt ctatatgggt ttcttaataa 1020
tggaattcag agagacttgc agttcttctt caacttttgt gatttccggt ctggggatga 1080
tgattatgaa acaatagcca tgtccacgat gcacacagat gtttccaaaa cttctttgaa 1140
gcaagcaagc ccagtcgcat ttaaaaaaat caacaacaat gatgataatg aaaaaatctg 1200
a / 1201

```

<210> 17

<211> 394

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Y5/Y1 CHIMERA

<400> 17

```

Met Ser Phe Tyr Ser Lys Gln Asp Tyr Asn Met Asp Leu Glu Leu Asp
  1           5           10           15

```

```

Glu Tyr Tyr Asn Lys Thr Leu Ala Thr Glu Asn Asn Thr Ala Ala Thr
      20           25           30

```

```

Arg Asn Ser Asp Phe Pro Val Trp Asp Asp Tyr Lys Ser Ser Val Asp
      35           40           45

```

```

Asp Leu Gln Tyr Phe Leu Ile Gly Leu Tyr Thr Phe Val Ser Leu Leu ---
  50           55           60

```

```

Gly Phe Met Gly Asn Leu Leu Ile Leu Met Ala Leu Met Lys Lys Arg
  65           70           75           80

```

```

Asn Gln Lys Thr Thr Val Asn Phe Leu Ile Gly Asn Leu Ala Phe Ser
      85           90           95

```

```

Asp Ile Leu Val Val Leu Phe Cys Ser Pro Phe Thr Leu Thr Ser Val
    100           105           110

```

Leu Leu Asp Gln Trp Met Phe Gly Lys Val Met Cys His Ile Met Pro
 115 120 125

Phe Leu Gln Cys Val Ser Val Leu Val Ser Thr Leu Ile Leu Ile Ser
 130 135 140

Ile Ala Ile Val Arg Tyr His Met Ile Lys His Pro Ile Ser Asn Asn
 145 150 155 160

Leu Thr Ala Asn His Gly Tyr Phe Leu Ile Ala Thr Val Trp Thr Leu
 165 170 175

Gly Phe Ala Ile Cys Ser Pro Leu Pro Val Phe His Ser Leu Val Glu
 180 185 190

Leu Gln Glu Thr Phe Gly Ser Ala Leu Leu Ser Ser Arg Tyr Leu Cys
 195 200 205

Val Glu Ser Trp Pro Ser Asp Ser Tyr Arg Ile Ala Phe Thr Ile Ser
 210 215 220

Leu Leu Leu Val Gln Tyr Ile Leu Pro Leu Val Cys Leu Thr Val Ser
 225 230 235 240

His Thr Ser Val Cys Ile Arg Leu Lys Arg Arg Asn Asn Met Met Asp
 245 250 255

Lys Met Arg Asp Asn Lys Tyr Arg Ser Ser Arg Ser Arg Ser Val Phe
 260 265 270

Tyr Arg Leu Thr Ile Leu Leu Ile Val Pro Ala Val Ser Trp Met Pro
 275 280 285

Leu His Leu Phe His Val Val Thr Ala Phe Asn Asp Asn Leu Ile Ser
 290 295 300

Asn Arg His Phe Lys Leu Val Tyr Cys Ile Cys His Leu Leu Gly Met
 305 310 315 320

Met Ser Cys Cys Leu Asn Pro Ile Leu Tyr Gly Phe Leu Asn Asn Gly
 325 330 335

Ile Gln Arg Asp Leu Gln Phe Phe Phe Asn Phe Cys Asp Phe Arg Ser
 340 345 350

Arg Asp Asp Asp Tyr Glu Thr Ile Ala Met Ser Thr Met His Thr Asp
 355 360 365

WO 03/104255

PCT/US02/33603

Val Ser Lys Thr Ser Leu Lys Gln Ala Ser Pro Val Ala Phe Lys Lys
370 375 380

Ile Asn Asn Asn Asp Asp Asn Glu Lys Ile
385 390